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Formation of Pyrophosphate, Tripolyphosphate, and Phosphorylimidazole With the Thioester, N, S-Diacetylcysteamine, As the Condensing Agent

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Summary. Reaction of 0.20M orthophosphate with 0.20M N,S-diacetylcysteamine in 0.40M imidazole at pH 7.0 or 8.0 under drying conditions at 50°C for 6 days yields pyrophosphate and tripolyphosphate in the presence and absence of 0.10M divalent metal ion. The efficiency of utilization of N,S-diacetylcysteamine in the formation of pyrophosphate linkages ranges from 3 – 8% under the above conditions. The thioester, N,S-diacetylcysteamine, and imidazole are required for phosphoanhydride formation.

Reaction of 0.40M orthophosphate with 0.20M N, S-diacetylcysteamine in 0.40M imidazole at ambient temperature for 6 days yields phosphorylimidazole in the absence or presence of 0.05M $MgCl_2$. Phosphorylimidazole and pyrophosphate are formed in the presence of 0.05M $CaCl_2$; pyrophosphate and tripolyphosphate are formed with 0.15M $CaCl_2$. The efficiency of utilization of N,S-diacetylcysteamine in the formation of pyrophosphate linkages is roughly 7% at 6 days of reaction with 0.15M $CaCl_2$. The thioester, N,S-diacetylcysteamine and imidazole are required for the formation of phosphoanhydrides. The significance of these reactions to molecular evolution is discussed.

Key words: Thioester – Pyrophosphate – Tripolyphosphate – Phosphorylimidazole – Molecular evolution – Prebiotic

Introduction

We previously reported the formation of the thioester, N,S-diacetylcysteine, by illumination of an aqueous solution of acetaldehyde and N, N'-diacetylcystine with ultraviolet light (Weber 1981). We now describe the formation of pyrophosphate, tripolyphosphate and phosphorylimidazole by using the thioester, N,S-diacetylcysteamine, as a condensing agent. Taken together these two reactions form a chemical pathway that may have provided the free energy needed for prebiotic biopolymer synthesis. This pathway resembles, in some respects, contemporary substrate-level oxidative phosphorylation reactions that use glyceraldehyde-3-phosphate, an α-keto acid or acetaldehyde as a substrate (Slater 1966; Goldman and Vagelos 1964; Harris and Waters 1976; Bridger 1974). These biochemical reactions proceed by the oxidation of an aldehyde to yield a thioester, which is used to drive the phosphorylation of a nucleoside diphosphate.

In his speculation on the autotrophic origin of metabolism Hartmann (1975) has stressed the importance of thioesters in prebiotic chemistry. Buvet (1977) has discussed the energetics of thioester formation as a prebiotic process for producing useful free energy. Amino acid thioesters also have been shown to condense efficiently to give peptides (Weber and Orgel 1979).

The formation of phosphoanhydrides is considered important to prebiotic chemistry because phosphonhydrides are plausible condensing agents for prebiotic biopolymer synthesis (Hulshof and Ponnamperuma 1976; Oro and Stephen-Sherwood 1976). Earlier investigations of prebiotic phosphoanhydride formation have used thermal methods (Rabinowitz et al. 1968; Osterberg and Orgel 1972; Handschuh et al. 1973) and chemical condensing agents, other than thioesters (Steinman et al. 1964; Miller and Parris 1964; Beck and Orgel 1965; Ferris 1968).

Abbreviations: P_1, orthophosphate; P_2, pyrophosphate; P_3, tripolyphosphate; ImP, phosphorylimidazole; Ac-Csa(Ac), N, S-diacetylcysteamine; Im, imidazole

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Experimental

Materials. N,S-diacetylcysteamine was obtained from Tridom Chemical Co., sodium phosphate (monobasic) from Mallinckrodt; sodium pyrophosphate from Baker Chemical Co.; sodium tripolyphosphate from Monsanto; imidazole from Sigma and carrier free sodium dihydrogen [^{33}P]-phosphate from New England Nuclear.

Chromatography. Paper Chromatography was carried out by descending elution on Whatman 3MM paper in System I with 95% ethanol, 1M ammonium acetate made up to 2 x 10^{-3} in EDTA and brought to pH 5.0 with glacial acetic acid (7:3 v/v); in System II with trichloroacetic acid (2.5g), dioxane (70ml), water (25ml), ammonium hydroxide (0.25 ml) and in System III with 95% ethanol, 0.1M potassium carbonate (6.5:3.5 v/v). Table 1 lists the chromatographic mobilities of the substances studied.

The products formed from radioactive [^{33}P]-orthophosphate were located and estimated by running the chromatograms through a Baird Atomic RSC-363 radioachromatogaphic scanner with entergratior. The reaction products were identified by co-chromatography with commercially available standards, whenever possible. The identification of phosphorylimidazole is supported by co-chromatography with authentic phosphorylimidazole, which was synthesised by the method of Lloyd and Cooperman (1971).

Reaction of N,S-diacetylcysteamine and Orthophosphate. In a typical reaction 30 µl of 1.0M sodium phosphate (pH 7.0), 30 µl of 1.0M imidazole · HCl (pH 7.0), 15 µl of water, 15 µl (approx. 1 µCi) of carrier free sodium dyhydrogen [^{33}P]-phosphate (50–1000mCi)mmol phosphorus), 30 µl of 0.5M magnesium chloride and 30 µl of 1.0M N,S-diacetylcysteamine were added in the order listed to a small test tube with vigorous agitation after each addition. In dry state reactions 20 µl aliquots of the reaction mixture were applied to 0.5 cm squares of Whatman GF 82 glass fiber paper. Except for the zero time sample, the squares were heated in open test tubes for various lengths of time at 50°C. Twenty microliters of 0.25M EDTA was immediately added to the square that held the zero time sample. At the end of each reaction 40 µl of 0.08M EDTA was added to the squares that were heated at 50°C. The glass fiber paper was then broken up with a glass rod and the resulting slurry was frozen at -70°C until analyzed. Reactions at ambient temperature in the presence of water were carried out in sealed test tubes without glass fiber paper and with a small amount of toluene added in order to prevent bacterial growth. The radioactive products were analyzed by chromatography and radiochromatographic scanning, as described earlier. The N,S-diacetylcysteamine was measured spectrophotometrically at its ϵ max at 233 nm (Noda et al. 1953).

Results

Phosphoanhydride Formation Under Drying Conditions

Fig. 1(a) shows the formation of pyrophosphate (P_2) and tripolyphosphate (P_3) by heating to dryness at 50°C reaction mixtures that contain orthophosphate (P_1), imidazole and N,S-diacetylcysteamine (Ac-Csa(Ac). Fig. 1(b) shows the same reaction in the presence of $MgCl_2$. The disappearance of Ac-Csa(Ac) is slower in the presence of $MgCl_2$. The final yield of P_2 is significantly

Table 1. Chromatographic Mobilities (Rm)[a]

Substance	System I Rm	System II Rm	System III Rm
P_1	1.00	1.00	1.00
P_2	0.55	0.56	0.47
P_3	0.30	0.31	0.47
ImP	0.77	–	1.74

a Chromatographic mobilities are given relative to P_1

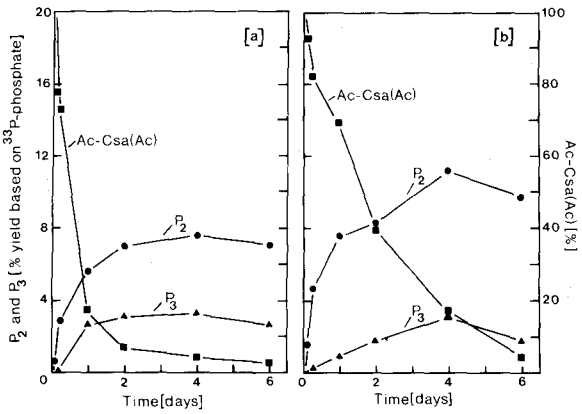


Fig. 1 a and b. Formation of pyrophosphate (P_2) and tripolyphosphate (P_3) by drying at 50°C reaction mixtures that contained 0.20M sodium [^{33}P]-phosphate (pH 8.0), 0.40M imidazole HCl (pH 8.0), 0.20M N,S-diacetylcysteamine [Ac-Csa(Ac)] (a) without divalent metal ion, (b) with 0.10M $MgCl_2$

Table 2. Formation of pyrophosphate and tripolyphosphate by heating at 50°C reaction mixtures that contained 0.20M sodium [^{33}P]-phosphate, 0.40M–1.0M imidazole, 0.20M N, S-diacetylcysteamine and 0.10M divalent metal ion

Buffer	Time (days)	Percent Yield Mg^{2+} P_2	P_3	Zn^{2+} P_2	P_3	Ca^{2+} P_2	P_3
0.4M Im(pH 7.0)							
	0.21	6.1	0.9	0.5	0.0	3.5	0.5
	1	6.8	1.5	1.7	0.5	5.7	1.0
	2	8.4	1.2	2.5	0.6	2.5	2.0
	4	9.8	3.0	2.9	1.4	9.5	3.1
	6	11.9	3.0	4.3	1.1	10.3	3.9
0.4M Im(pH 8.0)							
	0.21	5.7	0.3	1.5	0.2		
	1	8.5	1.4	5.0	0.3		
	2	10.5	1.4	11.3	0.9		
	4	10.8	2.1	12.4	2.3		
	6	9.6	1.3	13.1	1.7		
1.0M Im(pH 8.0)							
	0.21	2.5	0.4				
	1	2.6	0.2				
	2	2.5	0.0				
	4	3.2	0.0				

greater in the presence of $MgCl_2$, but the final yield of P_3 is slightly less in the presence of $MgCl_2$. P_2 and P_3 are not formed in measurable amounts (> 0.2%), when either the thioester, Ac-Csa(Ac), or imidazole are omitted from the reaction mixtures. In a control reaction without imidazole and Mg^{2+}, 85% of the Ac-Csa(Ac) remains at the end of the 6 days. In a similar reaction without imidazole in the presence of Mg^{2+} there is no measurable decrease in Ac-Csa(Ac).

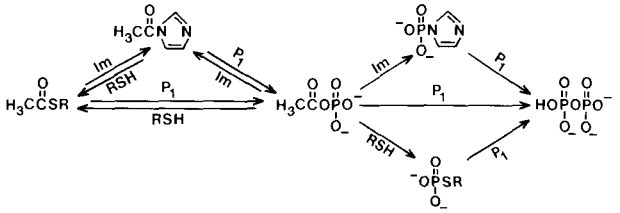
Table 2 shows the effect of metal ions, pH and imidazole concentration on the formation of P_2 and P_3 with the condensing agent, Ac-Csa(Ac). In 0.4M imidazole at pH 7.0 the final yields pf P_2 and P_3 are similar for Mg^{2+} and Ca^{2+} but much lower for Zn^{2+}. In 0.4M imidazole at pH 8.0, the final yield of P_2 with Zn^{2+} is greater than with Mg^{2+}, although the P_3 yields for Zn^{2+} and Mg^{2+} are comparable. The yields of P_3 are greater at pH 7.0 compared to pH 8.0 in reactions with Mg^{2+}. The yields of P_2 and P_3 decrease dramatically when the imidazole concentration at pH 8.0 is increased from 0.4M to 1.0M in the presence of Mg^{2+}.

Phosphoanhydride Formation in the Presence of Water

Fig. 2(a – c) depicts the formation of P_2, P_3 and ImP in the presence and absence of Ca^{2+} in reaction mixtures that contain P_1, imidazole and Ac-Csa(Ac) at ambient temperature. Precipitates are present in reaction mixtures that contain divalent cations. Fig 2(a) shows that in the absence of a divalent cation the only phosphoanhydride product is ImP. Similar reactions with 0.20M sodium phophate and 0.05M $MgCl_2$ at pH 7.0 and 8.0 yield, respectively, 0.8% ImP and 1.5% ImP, each with negligible P_2. However, as shown in Fig. 2(b), the presence of 0.05M Ca^{2+} in the reaction mixture results in the formation of both ImP and P_2. Fig. 2(c) shows that 0.15M Ca^{2+} gives a higher yield of P_2 and a trace of P_3, but no measureable ImP. The yield of P_2 at 6 days in the presence of 0.30M Ca^{2+} is 1.3% with no measurable P_3 or ImP. Phosphoanhydrides are not formed in measurable amounts (> 0.2%) in reactions similar to that in Fig. 2(b) that do not contain imidazole or Ac-Csa(Ac) or that have 2,6-lutidine substituted for imidazole. The Ac-Csa(Ac) that remains after 6 days of reaction is 95% when imidazole is absent, and 90% when 2,6-lutidine is substituted for imidazole.

Discussion

We have shown that the thioester, Ac-Csa(Ac), can be used to drive the synthesis of (1)P_2 and P_3 under drying conditions at 50°C, (2) ImP at ambient temperature in the presence of water, and (3) P_2, P_3 and ImP at ambient temperature in the presence of Ca^{2+} and water. As shown in the scheme below, the formation of P_2, P_3 and ImP is thought to occur via acetyl phosphate as an intermediate.



Acetyl phosphate is produced by reaction of acetyl thioester with P_1, or by reaction of acetyl thioester with imidazole to give acetylimidazole, which subsequently reacts with P_1 (Bruice et al. 1963; Stadtman 1954; Heller et al. 1977). Acetyl phosphate goes on to react with (1) P_1 to form P_2 directly (DiSabato and Jencks 1961b; Etaix and Buvet 1975), (2) imidazole to give ImP, which subsequently reacts with P_1 to yield P_2 (Gibbs et al. 1980), or (3) thiol to yield a phosphorothioate, which reacts with P_1, to give P_2 (Dittmer and Silverstein 1961). Further phosphorylation of P_2 by similar reactions yields P_3.

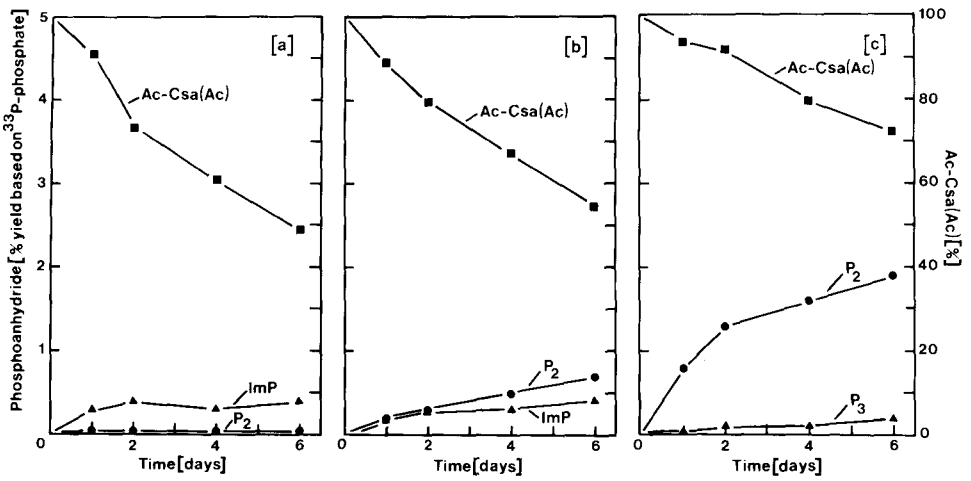


Fig. 2 a–c. Formation of pyrophosphate (P_2) and tripolyphosphate (P_3) and phosphorylimidazole (ImP) at ambient temperature in reaction mixtures that contained 0.20M sodium [^{33}P]-phosphate (pH 7.0), 0.40M imidazole · HCl (pH 8.0), 0.10M N, S-diacetylcysteamine [Ac-Csa(Ac)] **(a)** without divalent metal ion, **(b)** with 0.05M $CaCl_2$ and **(c)** with 0.15M $CaCl_2$

The major obstacle to efficient formation of $P_2(P_3)$ is hydrolysis of acetyl thioester and reaction intermediates. The very low yield (0.3%) of P_2 from 0.2M acetyl phosphate (Etaix and Buvet 1975) suggests that much of the energy loss in converting thioester to P_2 (P_3) occurs by hydrolysis of acetyl phosphate (Koshland 1952; DiSabato and Jencks 1961a,b; Oestreich and Jones 1966), although losses due to hydrolysis of the acetyl thioester (Hawkins and Tarbell 1953; Weber 1981), acetylimidazole (Jencks and Carriulo 1959a,b), phosphorylimidazole (Jencks and Gilchrist 1965; Lloyd and Cooperman 1971) and phosphorothioate (Dittmer et al. 1963) could be significant. Losses due to hydrolysis of P_2 and P_3 are negligible, since these substances are very stable (Wazer et al. 1955).

Imidazole apparently is acting as a catalyst in $P_2(P_3)$ formation, since control reactions without imidazole or with 2,6-lutidine substituted for imidazole do not yield $P_2(P_3)$. The formation of ImP in reactions carried out in the presence of water without Ca^{2+}, suggests that the catalytic role of imidazole involves formation of ImP, which subsequently phosphorylates P_1 to give P_2. The actual amounts of ImP that form in these experiments is greater than the measured values because ImP hydrolyzes during the reactions ($t_{1/2}$ = 960 min at 39°C, pH 7, Jencks and Gilchrist 1965). Although it is likely that ImP forms in reactions under drying conditions at 50°C, reaction of ImP with the concentrated P_1 to give P_2 would prevent the accumulation of ImP. Likewise, the absence of ImP in reactions in the presence of water and 0.15M Ca^{2+} could be accounted for by reaction of the ImP with P_1 on the surface of the calcium phosphate precipitate. This contention is supported by the demonstration that adenosine 5-phosphorimidazolide reacts with P_1 of hydroxyapatite to give ADP (Gibbs et al. 1980).

In the 0.4M imidazole buffer, the efficiency of utilization of thioester in the formation of pyrophosphate linkages ranges from 3 – 8% under drying conditions at 50°C with and without divalent metal cations. However, the formation of pyrophosphate linkages in water at ambient temperature occurs after 6 days of reaction with a moderate efficiency (7%) only in the presence of 0.15M Ca^{2+}. The effect of Ca^{2+} on $P_2(P_3)$ formation probably involves the reaction of precipitated calcium phosphate with one or more 'energy-rich' substances in the proposed reaction scheme. Earlier studies of P_2 formation with cyanate as the condensing agent have shown a similar dependence for an insoluble calcium phosphate phase, hydroxyapatite (Miller and Parris 1964; Beck and Orgel 1965). Calcium phosphate precipitates and hydroxyapatite have also been shown to participate in the transphosphorylation of P_1 by adenosine di- and triphosphates to give P_2 (Burley 1965; Krane and Glimcher 1962). Etaix and Buvet (1975) have reported that Ca^{2+} catalyzes the transphosphorylation of P_1 by P_3 to give P_2.

Our earlier studies on the photochemical production of a thioester from a disulfide and acetaldehyde (Weber 1981), together with this demonstration that thioesters can drive the formation of phosphoanhydrides, describes a pathway for the generation of 'energy-rich' phosphoanhydrides in the aqueous environment of the prebiotic Earth. Since the concentration of P_1 in the prebiotic oceans was very low (Miller and Parris 1964, Griffith et al. 1977), we presume that the formation of thioesters and their use in $P_2(P_3)$ synthesis occured with thiols (disulfides) that were adsorbed to the surface of a phosphate mineral, like hydroxyapatite. A thiol (disulfide) in a polyanionic peptide or nucleotide derivative, like coenzyme A, would be expected to bind to hydroxyapatite (Bernardi and Kawasaki 1968; Bernardi 1971; Burton et al 1969). Alternatively, imidazole residues of substances, like histidine-containing polyanionic peptides that adhere to hydroxyapatite, may have efficiently catalyzed $P_2(P_3)$ formation from thioesters formed in solution.

Phosphoanhydrides formed from thioesters in the prebiotic environment would provide the free energy necessary for biopolymer synthesis. Phosphoanhydrides that have been used to form peptide bonds under presumed prebiotic conditions include pyrophosphate (Rabinowitz et al. 1969), tripolyphosphate (Rabinowitz et al. 1969), trimetaphosphate (Chung et al. 1971), polyphosphate (Harada and Fox 1965; Rabinowitz et al. 1969); adenosine triphosphate (Sawai et al. 1975; Weber et al. 1977; Nakashima and Fox 1980) and adenosine 5'-phosphorimidazolide (Lohrmann et al. 1975). Phosphoanhydrides have also been used in the prebiotic synthesis of nucleotides (Ponnamperuma 1963; Schwartz and Ponnamperuma 1968; Lohrmann 1976; Etaix and Orgel 1978), nucleoside phosphoamidates (Lohrmann 1977 and Sleeper et al. 1978), and oligonucleotides (Schwartz and Fox 1967; Weber et al. 1977; Lohrmann and Orgel 1978).

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References

Beck A, Orgel LE (1965) The formation of condensed phosphate in aqueous solution. Proc Natl Acad Sci USA 54:664–667

Bernardi G (1971) Chromatography of proteins on hydroxyapatite. In Colowick SP, Kaplan NO (eds) Methods in enzymology, vol XXII. Academic Press, New York, p 325

Bernardi G, Kawasaki T (1968) Chromatography of polypeptides and proteins on hydroxyapatite columns. Biochim Biophys Acta 160:301–310

Bridger WA (1974) Succinyl-CoA synthetase. In: Boyer PD (ed) The enzymes, vol X. Academic Press, New York, p 581

Bruice TC, Bruno JJ, Chou W (1963) Nucleophilic displacement reactions at the thiol-ester bond of δ-thiovalerolactone. J Am Chem Soc 85:1659–1669

Burley RW (1965) Transphosphorylation of adenosine di- and tri-phosphates in the presence of calcium phosphate precipitates. Nature 208:683–684

Burton FG, Neuman MW, Neuman WF (1969) On the possible role of crystals in the origins of life. I. The adsorption of nucleosides, nucleotides and pyrophosphate by apatite crystals. Curr Mod Biol 3:20–26

Buvet R (1979) Compared energetics of primordial and biolocical metabolisms. Origins of Life 8:361–370

Chung NM, Lohrmann R, Orgel LE (1971) The mechanism of the trimetaphosphate induced peptide synthesis. Tetrahedron 27:1205–1210

DiSabato G, Jencks WP (1961a) Mechnism and catalysis of reactions of acyl phosphates. I. Nucleophilic reactions. J Am Chem Soc 83:4393–4400

DiSabato G, Jencks WP (1961b) Mechanism and catalysis of reactions of acyl phosphates. II. Hydrolysis. J Am Chem Soc 83:4400–4405

Dittmer DC, Ramsay OB, Spalding RE (1963) Reactivity of thiophosphates. II. Hydrolysis of S-n-butylphosphorothioate and S-(2-aminoethyl)phosphorothioate. J Org Chem 28:1273–1278

Dittmer DC, Silverstein VO (1961) Production of pyrophosphate from S-n-butylphosphorothioate. J Org Chem 26:4706–4707

Etaix E, Buvet R (1975) Conditions of occurrence for primeval processes of transphosphorylations. Origins of Life 6:175–183

Etaix E, Orgel LE (1978) Phosphorylation of nucleosides in aqueous solution using trimetaphosphate: Formation of nucleoside triphosphates. J Carbohydr Nucleosides Nucleotides 5:91–110

Ferris JP (1968) Cyanovinyl phosphate: A prebiological phosphorylating agent? Science 161:53–54

Gibbs D, Lohrmann R, Orgel LE (1980) Template-directed synthesis and selective adsorption of oligoadenylates on hydroxyapatite. J Mol Evol 15:347–354

Goldman P, Vagelos PR (1964) Acyl-transfer reactions (CoA-structure, function). In: Florkin M, Stotz EH (eds) Comprehensive biochemistry, vol 15. Elsevier, Amsterdam, p 71

Griffith EJ, Ponnamperuma C, Gabel NW (1977) Phosphorus, a key to life on the primitive earth. Origins of Life 8:71–85

Handschuh GJ, Lohrmann R, Orgel LE (1973) The effect of Mg^{2+} and Ca^{2+} on urea-catalyzed phosphorylation reactions. J Mol Evol 2:251–262

Harada K, Fox SW (1965) Thermal polycondensation of free amino acids with polyphosphoric acid. In: Fox SW (ed) The origins of prebiological systems and of their molecular matrices. Academic Press, New York, p 289

Harris JI, Waters M (1976) Glyceraldehyde-3-phosphate dehydrogenase. In: Boyer PD (ed) The enzymes, vol XIII. Academic Press, New York, p 1

Hartman H (1975) Speculations on the origin and evolution of metabolism. J Mol Evol 4:359–370

Hawkins PJ, Tarbell DS (1953) Studies on model compounds for coenzyme A. A kinetic study of aminolysis and hydrolysis of ethyl thioacetate and β-acetaminoethyl thioacetate in aqueous solution. J Am Chem Soc 75:2982–2985

Heller MJ, Walder JA, Klotz IM (1977) Intramolecular catalysis of acylation and deacylation in peptides containing cysteine and histidine. J Am Chem Soc 99:2780–2785

Hulshof J, Ponnamperuma C (1976) Prebiotic condensation reactions in an aqueous medium: A review of condensing agents. Origins of Life 7:197–224

Jencks WP, Carriuolo J (1959a) Imidazole catalysis: II. Acyl transfer and the reactions of acetyl imidazole with water and oxygen anions. J Biol Chem 234:1272–1279

Jencks WP, Carriuolo J (1959b) Imidazole catalysis III. General base catalysis and the reactions of acetyl imidazole with thiols and amines. J Biol Chem 234:1280–1285

Jencks WP, Gilchrist M (1965) Reactions of nucleophilic reagents with phosphoramidate. J Am Chem Soc 87:3199–3209

Koshland DE (1952) Effect of catalysts on the hydrolysis of acetyl phosphate. Nucleophilic displacement mechanisms in enzymatic reactions. J Am Chem Soc 74:2286–2292

Krane SM, Glimcher MJ (1962) Transphosphorylation from nucleoside di- and triphosphates by apatite crystals. J Biol Chem 237:2991–2998

Loyd GJ, Cooperman BS (1971) Nucleophilic attack by zinc (II)-pyridine-2-carbaldoxime anion on phosphorylimidazole. A model for enzymatic phosphate transfer. J Am Chem Soc 93:4883–4889

Lohrmann R (1976) Formation of nucleoside 5'-polyphosphates under potentially prebiological conditions. J Mol Evol 8:197–210

Lohrmann R (1977) Formation of nucleoside 5-phosphoramidates under potentially prebiological conditions. J Mol Evol 10:137–154

Lohrmann R, Orgel LE (1978) Template-directed polynucleotide condensation as model for RNA replication. In: Noda H (ed) Origin of Life. Japan Scientific Societies Press, Tokyo, p 235

Lohrmann R, Ranganathan R, Sawai H, Orgel LE (1975) Prebiotic peptide-formation in the solid state. I. Reactions of benzoate ion and glycine with adenosine 5'-phosphorimidazolide. J Mol Evol 5:57–73

Miller SL, Parris M (1964) Synthesis of pyrophosphate under primitive earth conditions. Nature 204:1248–1250

Nakashima T, Fox SW (1980) Synthesis of peptides from amino acids and ATP with lysine-rich proteinoid. J Mol Evol 15: 161–168

Noda LH, Kuby SA, Lardy HA (1953) Properties of thioesters: Kinetics of hydrolysis in dilute aqueous media. J Am Chem Soc 75:913–917

Oestreich CH, Jones MM (1966) The effect of metal ions on labile phosphates. I. The hydrolysis of acetyl phosphate dianion. Biochemistry 5:2926–2931

Oro J, Stephen-Sherwood E (1976) Abiotic origin of biopolymers. Origins of Life 7:37–47

Osterberg R, Orgel LE (1972) Polyphosphate and trimetaphosphate formation under potentially prebiotic conditions. J Mol Evol 1:241–248

Ponnamperuma C, Sagan C, Mariner R (1963) Synthesis of adenosine triphosphate under possible primitive earth conditions. Nature 199:222–226

Rabinowitz J, Chang S, Ponnamperuma C (1968) Phosphorylation on the primitive earth: Phosphorylation by way of inorganic phosphate as a potential prebiotic process. Nature 218:442–443

Rabinowitz J, Flores J, Krebsbach R, Rogers G (1969) Peptide formation in the presence of linear or cyclic polyphosphates. Nature 224:795–796

Sawai H, Lohrmann R, Orgel LE (1975) Prebiotic peptide-formation in the solid state. II. Reaction of glycine with adenosine 5'-triphosphate and P_1, P_2-diadenosine-pyrophosphate. J Mol Evol 6:165–184

Schwartz AW, Fox SW (1967) Condensation of cytidylic acid in the presence of polyphosphoric acid. Biochim Biophys Acta 134:9–16

Schwartz A, Ponnamperuma C (1968) Phosphorylation on the primitive earth: Phosphorylation of adenosine with linear polyphosphate salts in aqueous solution. Nature 218: 443

Slater EC (1966) Oxidative phosphorylation. In: Florkin M, Stotz EH (eds) Comprehensive biochemistry, vol 14. Elsevier, Amsterdam p 327

Sleeper HL, Lohrmann R, Orgel LE (1978) Formation of the imidazolides of dinucleotides under potentially prebiotic conditions. J Mol Evol 11:87–93

Stadtman ER (1954) On the energy-rich nature of acetyl imidazole, an enzymatically active compound. In: McElroy WD, Glass B (eds) The mechanism of enzyme action. John Hopkins Press, Baltimore, p 581

Steinman G, Lemmon RM, Calvin M (1964) Cyanamide: A possible key compound in chemical evolution. Proc Natl Acad Sci USA 52:27–30

Wazer JRV, Griffith EJ, McCullough JF (1955) Structure and properties of the condensed phosphates. VII. Hydrolytic degradation of pyro- and tripolyphosphate. J Am Chem Soc 77:287–291

Weber AL, Caroon JM, Warden JT, Lemmon RM, Calvin M (1977) Simultaneous peptide and oligonucleotide formation in mixtures of amino acid, nucleoside triphosphate, imidazole, and magnesium ion. BioSystems 8:277–286

Weber AL, Orgel LE (1979) The formation of peptides from glycine thioesters. J Mol Evol 13:193–202

Weber AL (1981) Formation of the thioester, N, S-diacetylcysteine, from acetaldehyde and N,N'-diacetylcystine in aqueous solution with ultraviolet light. J Mol Evol 17:103–107

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