

PHOSPHORYLATION AND OLIGOMERIZATION OF NUCLEOSIDES IN A UREA-RICH EUTECTIC SOLVENT USING MINERAL AND FREE PHOSPHATE SOURCES. B. T. Burcar¹, M. Pasek², M. Gull², B. J. Cafferty¹, F. Velasco³, N. V. Hud¹, and C. Menor-Salvan¹, ¹School of Chemistry and Biochemistry, Georgia Institute of Technology, Atlanta, GA 30033 (USA), ²School of Geosciences, University of South Florida, Tampa, FL (USA), ³Dept. of Mineralogy and Petrology, Universidad del Pais Vasco, Campus de Leioa, Vizcaya, Spain.

Introduction: Phosphate plays a vital role in modern biology by ensuring the solubility of DNA, RNA, and a range of metabolic compounds while providing moderately stable phosphodiester bonds which are able to be readily formed and cleaved by modern enzymes. On a prebiotic Earth these properties would also have been invaluable for the organic molecules that gave rise to the origins of life. Life almost certainly arose in aqueous environments making solubility in water under varied pH conditions quite important, a property readily observed by phosphorylated organic molecules. Equally important is the ability of nascent life to form informational carrying polymers with chemical bonds that are easily formed and strong enough to preserve informational fidelity, but are weak enough to allow the recycling of the monomeric units for chemical reuse and the continued sampling of different informational sequences. This thermodynamic sweet spot is found with phosphorylated organics in modern biochemistry and is one of the main reasons phosphate is such a fundamental part of all living systems.

These properties would have made phosphate an important molecule and moiety to be harnessed in early prebiotic chemical reactions. In fact, early work has shown the ability of urea-catalyzed phosphorylation to be utilized for phosphorylating alcohol containing organics at moderate temperatures.¹

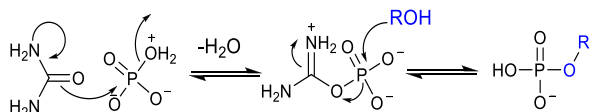


Fig. 1. Proposed mechanism for urea-catalyzed synthesis of phosphate esters.²

However, on a prebiotic Earth, phosphate would have readily reacted with divalent cations in solution (primarily magnesium, iron, and calcium) to form insoluble precipitates which would have prevented phosphate from being incorporated into developing biochemical systems. In addition, the phosphorylation of nucleosides is a dehydration reaction making these reactions thermodynamically unfavorable in aqueous environments. These two factors create sizable barriers for phosphate to be utilized by nascent life.

Results and Discussion: Our work builds upon previous work demonstrating successful phosphorylation in eutectic solvents,³ investigating urea-catalyzed phosphorylation in a prebiotically viable eutectic solvent consisting of urea, ammonium formate, and water (UAFW Eutectic). In this solution, high degrees of phosphorylation were observed with ribonucleosides or glycerol when evaporatively heated in the presence of free phosphate in solution. In addition, in the presence of $MgSO_4$, water insoluble hydroxyapatite ($Ca_{10}(PO_4)_6(OH)_2$) was able to be successfully utilized as a phosphorylation source. These results demonstrate the ability of the UAFW eutectic to not only access urea-catalyzed phosphorylation in a liquid environment, but to mobilize previously sequestered phosphate for these reactions.⁴ This work shows a prebiotic chemical path to phosphorylation of nucleosides which overcomes two of the major barriers to phosphorylation, helping support phosphate as an important molecule utilized during the origins of life.

In addition, upon extended heating of these solutions (7-30 days depending on temperature), the formation of phosphodiester bonds were observed. Dimers and trimers were formed containing either adenosine monophosphate or glycerol phosphate subunits.⁴ These exciting results demonstrate the potential of not only utilizing urea-catalyzed phosphorylation to make organophosphates, but also using this reaction chemistry to make longer biopolymers under prebiotic conditions.

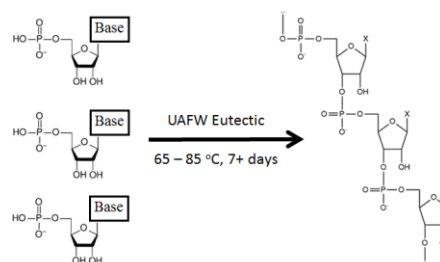


Fig. 2. UAFW driven polymerization from nucleotides.

References: [1] Österberg R., Orgel L., Lohrmann R. J. (1973) *Mol. Evol.* 2, 231–234. [2] Steinman G., Lemmon R. M., Calvin M. (1964) *Proc. Natl. Acad. Sci. USA* 52, 27–30. [3] Gull M., et al. (2014) *J. Mol. Evol.* 78, 109–117. [4] Burcar B. et al. (2016) *Angew. Chem. Int. Ed.* 55, 13249–13253.