Illuminating Life’s Origins: UV Photochemistry in Abiotic Synthesis of Biomolecules

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1. INTRODUCTION
Attempts to understand the provenance of life’s building blocks on Earth have led to the study of the innate reactivity of diverse planetary chemical feedstocks. The question of how simple molecules available on the primordial Earth may have interacted to build systems of increasing complexity can be probed by considering the environmental inputs of a nascent Earth and coupling them with material feedstocks to develop scenarios in which molecules of interest—implicated by their role in modern biology—are formed. Refinement of these scenarios is best served by a two-way discourse, by which we consider our knowledge of chemical reactivity and how it might lead us to important biomolecules, but also by which we confine ourselves to plausible proposed geochemical scenarios and attempt to discover new chemistries within them to achieve our chemical ends. In this way our group and others have considered potentially fruitful chemistries and their geochemical implications, and conversely explored the reaction networks of proposed geochemical scenarios. Many proposed environmental inputs have presented themselves as potentially useful, but a striking finding unifying much research in prebiotic synthesis is the role of light in promoting useful or otherwise difficult transformations, and presenting sometimes unexpected means by which important biomolecules are selectively synthesized. Light is indispensable in both the modern biological ecosystem, and, it would appear, laboratory emulations of abiotic chemical systems that are its ancient progenitor. Herein we discuss the major findings of the role of light in prebiotic synthesis, the implications for defining plausible prebiotic chemical synthesis/degradation scenarios, and the emerging future direction that this research is taking us. Given the nature of this overview, rather than exhaustively explore the role of photons in prebiotic synthesis, we focus somewhat on the work of our own laboratory, in which over the past decade photochemistry has emerged as a prominent theme, alongside the most significant other contributions.

Other reviews less focused on the role of light in prebiotic chemistry have been published in the past few years.1−5

While our work has repeatedly reminded us that discovering chemistry leading to biology is not served by splitting target molecules and reactions into groups, but by embracing systems chemistry, for ease of engagement this Perspective is divided into five sections before our conclusions and outlook. First, we briefly discuss historic prebiotic photochemistry in order to contrast with the following sections, which describe photochemistry mostly related to carbohydrate and amino acid derivative synthesis; the role of photochemistry in nucleoside synthesis; how photochemistry has enabled condensation of biomolecules; and photochemical selection. We conclude by discussing some implications of photochemistry on stability, selectivity, and plausibility, and the future directions of origins research. Given the ever increasing complexity of the systems approach in prebiotic chemistry, some overlap is unavoidable. We hope that, as the reader progresses, the superficiality of this classification and the inevitable entanglement of the abiotic synthetic chemistry of these classes of molecule becomes increasingly evident, along with the role of photochemistry in linking them.

2. HISTORICAL PERSPECTIVE
Light from the Sun has long been recognized as the principal source of energy to Earth. The amount of ultraviolet (UV) radiation incident on the young Earth’s surface would have been dependent on atmospheric composition, which is the
subject of some debate. In the absence of any haze, UV flux at the surface was likely orders of magnitude higher than it is today in the spectral range down to 200 nm, largely due to the absence of ozone. Ultraviolet light, lightning, vulcanism, and impactors were identified early as the most important geophysical driving forces of chemical reactions with the potential to generate organic matter. In Miller’s reports of his famous reducing atmosphere electric discharge experiments, he explicitly considered UV irradiation as an alternative to electric discharge, but decided that the latter would be easier to probe experimentally and would be of more relevance in generating organic matter relatively close to the terrestrial surface, where transport to the ocean could occur before photolytic degradation. Hundreds of (largely less successful) Urey–Miller-type experiments in atmospheres of various compositions, using UV as energy input, were subsequently reported. However, little headway was made into improving the selectivity of Miller’s experiments or synthesizing biomolecules more complex than amino acids (e.g., nucleosides). Here we draw the distinction between atmospheric photochemistry and surficial aqueous photochemistry. The former, alongside chemistry promoted by electric discharge, vulcanism, and impactors would have shaped the structurally simple (volatile) constituency of the prebiotic Earth’s atmosphere and, by equilibrium, oceans, mostly via photolysis. The latter has been shown to have the potential to drive the assembly of these simple feedstocks into chemical systems of greater structural and compositional complexity, highly suggestive of biological antecedence. The critical difference is that in the atmosphere, the energy of photons mostly results in the breakdown of organic matter, whereas in the more concentrated aqueous environment, irradiation can generate energetic species able to react in intermolecular fashion, or act on products of aqueous or solid state intermolecular chemistry. The selective increase in complexity afforded by surficial aqueous photochemistry is the focus of this Perspective. Whether such reactivity occurred on the young Earth would have depended on a variety of environmental factors, and a balance between photochemical synthesis and destruction. Defining what is the extent of chemical possibility in this regard has led to many discoveries in photochemistry, that broadly inform our understanding of the fundamental reactivity of (pre)biologically important molecules.

The past decade has seen noteworthy advances in the discovery of photochemistry in prebiotically relevant transformations and syntheses of (proto)biological molecules, which we will discuss in detail. However, a cornerstone of prebiotic chemistry, and a highly instructive example of the nuances of (photo)chemistry in origins of life studies, is the investigation by Ferris, Sanchez, and Orgel in the 1960s, of the thermal and photochemical pentamerization of hydrogen cyanide (HCN, 1) to adenine. The formation of adenine from ammoniacal cyanide, first discovered by Oro and subsequently repeated under numerous conditions of varying prebiotic relevance, is so celebrated that consideration of the physical and geochemical restrictions for this chemistry is sometimes neglected. The mechanism and outcome of HCN polymerization, and hence the yield of adenine, are highly dependent on the conditions and additives employed. Orgel et al. carefully investigated adenine formation under aqueous and eutectic conditions, at varying concentrations and with various additives, both in the dark and under UV irradiation, ultimately concluding that “the photochemical pathway involving only cyanide presents so few difficulties that we believe it must have been significant” (Scheme 1). This conclusion rested on thorough determination of the outcome and rates of polymerization and degradation pathways, and also the discovery of new chemistry—a marvelously facile and mechanistically intriguing photoisomerization of diamononitrile (DAMN, 2) to 5-aminimidazole-4-carbonitrile (AICN, 9), at 350 nm, which they found was readily converted to adenine and other purines. Their investigations confirmed that, although AICN 9 and adenine 10 are obtainable in low yields, the major tractable product of HCN oligomerization at reasonably plausible concentrations (0.01−0.1 M) is the HCN tetramer, diamononitrile (DAMN, 2) (Scheme 1). The same is true of eutectic-phase oligomerizations which give even greater yields of DAMN. DAMN may be thermally converted to AICN 9 or adenine 10 itself (Scheme 1, gray pathways), but for these reactions to compete with the measured rates of hydrolysis and degradation of the reactants, concentrations must be very high. For example, to form formamidine (HC(NH)NH2, 8) in reasonable steady state concentration from cyanide requires high concentrations of ammonia, hence adenine is synthesized in relatively high yield (15%) from cyanide in liquid ammonia, but, even in 15 N aqueous ammonia, only 0.5% adenine can be obtained. In comparison, AICN 9 is the most hydrolytically stable intermediate in the
various pathways to adenine, and forms near quantitatively from DAMN 2 after an afternoon of irradiation in bright sunlight. The investigators did note, however, that although pure AICN 9 is photochemically robust, at moderate

concentration its photochemical decomposition is accelerated by byproducts of HCN 1 polymerization, so that AICN 9 must either be protected from excessive irradiation or formed in highly dilute solution. Thus, the authors surmised that photochemical conversion of DAMN 2, formed by cyanide oligomerization, to the relatively stable AICN 9 followed by its conversion to adenine 10 or guanine, is probably the most plausible pathway to purine nucleobases, and could have occurred seasonally as lakes froze and thawed, or in falling raindrops. This work paved the way for following studies on the prebiotically plausible synthesis of adenine 10,31–32 and sets a benchmark for all prebiotic investigations in its thorough evaluation of plausibility through synthetic, kinetic, and systems chemical considerations. Clearly, to build a plausible geochemical scenario including radiative inputs, careful and thorough analysis of both photochemical and thermal processes, and potential competition or synergy between them, must be evaluated. The progress in this area, including not only constructive processes focusing on controlled radiative input, but also the potential destructiveness of irradiation, are discussed in this Perspective.

The mechanism of this photoisomerization was the subject of debate26–30 for decades after its report in 1966, but by 201331,32 combined theory and experiment concluded that the reaction is likely a process involving three photochemical steps (Scheme 1), due to constraints in the hot ground state imposed by ultrafast energy dissipation. Barbetti et al. calculated that after photoisomerization of DAMN 2 to its (E)-isomer 3 (in the triplet manifold), a second photoexcitation is required to promote hydrogen atom shift, forming spectroscopically detectable ketenimine 4. A third photon absorption promotes singlet manifold photocyclization, generating equilibrating excited state azetenes 7 and 6. In the excited state, 6 is able to undergo C–C bond cleavage and ultimate rearrangement to N-heterocyclic carbene 5, which tautomerizes readily to AICN, 9.

3. PHOTOREDUCTIVE HOMOLOGATION OF HCN AND OTHER NITRILES

Hydrogen cyanide is one of the simplest and most readily available planetary carbon feedstocks that can be converted into organic and biological molecules.33–35 Our laboratory discovered the propensity of HCN to undergo photoreductive homologation to generate carbohydrates in a prebiotic context.36–40 Over the past decade, we have demonstrated that this reductive homologation process results in the conversion of HCN and simple building blocks into a “cyanosulfidic” network of biologically relevant products. Our interest in purine synthesis and the beautiful and landmark cyanide oligomerization chemistry of Oró, Orgel, and others, combined with geochemical implications linking cyanide and various metals,7 led us to investigate reports of photochemical, copper-mediated oxidative coupling of cyanide proceeding with concomitant production of hydrated electrons.41,42 We thus irradiated with ultraviolet light (254 nm) a solution of HCN and copper(1) cyanide at near neutral pH and found that a photochemical redox cycle of cyanocuprates (Scheme 2, top left) was indeed an effective way of generating a reducing agent presumed to be hydrated electrons, produced by photodetachment from cyanocuprates.41 Although no purines were detected in our experiments, we recognized the operation of a prebiotic variant of the Kiliani-Fischer carbohydrate homologation sequence,43–45 which had the potential to provide a cleaner source of carbohydrates for nucleoside synthesis (see Section 4) than the notoriously unselective formose reaction.46 We rationalized the products formed by assuming that photochemically generated hydrated electrons (or hydrogen atoms which can be produced by protonation of hydrated electrons by general acids) first reduce HCN to formaldehyde imine 14 (Scheme 2). HCN 1 itself is the stoichiometric reductant, being oxidized to cyanogen 11 in the disproportionation of 1. The reduced product 14 is either converted to glyoxal nitrile 19, a precursor of glycine, by addition of excess of HCN 1, or hydrolyzed to formaldehyde 15 and converted by addition of cyanide 1 to glycolonitrile 16. A second, iterative round of reduction, hydrolysis, and addition affords glycolaldehyde imine 17, then glycolaldehyde 23, and its cyanohydrin 24. Imine 17 can also be converted to serine nitrile 18, another amino acid precursor, by addition of HCN 1 before hydrolysis. The third stage reduction of 24 by hydrated electrons, followed by hydrolysis, produces an additional homologated sugar, glyceraldehyde 31. Although we initially considered the formation of cyanogen 11 in a positive light given its potential role in the accelerated formation of purine precursors from HCN,15 its hydrolysis product, hydrogen cyanate 22, traps (probably irreversibly) both glycolaldehyde 23 and glyceraldehyde 31 as their cyclic cyanate adducts 13, precluding their function in a system that might ultimately produce nucleotides or other carbohydrate-containing biomolecules. Thus, while we had discovered interesting and promising reductive homologation chemistry, we needed to refine it.

Prompted by further considerations of a geochemical scenario that would facilitate a photooxidation cycle without generating cyanogen 11, we investigated a modified cycle of copper and cyanide featuring H2S as the ultimate reductant37 (Scheme 2, top middle). In the geological scenario, H2S and catalyst CuCN could be produced by dissolution of copper sulfide minerals in cyanide solution.47,48 H2S acts as the stoichiometric reductant to reduce Cu(II) complexes generated by photodetachment of an electron from Cu(I) complexes and complete the catalytic cycle. This occurs either thermally or by photodetachment of an electron from hydroxysulfide that performs the reduction, and thus H2S is oxidized to HS−/S2−, which likely reacts further to form H2S2/HSS−. Thiocyanate, which could form by reaction of cyanide with H2S2/HSS−, was detected as a byproduct, which our later work suggests is important (see Section 4). Apart from the production of the simple sugars 23 and 31, and the Strecker40 precursors (19 and 18) of glycine and serine, deoxygenated product acetaldehyde 29 was also detected. In separate experiments thiocyanate was found to be instrumental in the deoxygenation of 23. Strecker amino acid synthesis conditions convert 29 into the corresponding cyanoacrylic 28 and alanine precursor aminonitrite 27. Simultaneous photoreduction of 28 leads to lactaldehyde 34, which produced threonine precursor aminonitrite 33 by addition of 1 and NH3.

The coexistence of α-aminonitrite Strecker precursors of the amino acids glycine, serine, alanine, and threonine with carbohydrate ribonucleoside building blocks in the same photochemical scenario prompted further exploration.40 The interconversion of 31 and its more stable isomer, dihydroxyacetone 38, is promoted, by general acid–base catalysis, by phosphate. When 38 was subjected to photooxidation conditions, the deoxygenated product acetone 37 was produced alongside another reduced product, biological
membrane precursor glycerol. Addition of HCN to acetone 37 forms cyanohydrin 44. Photoreduction of 44 does not proceed efficiently because 44 is only a minor component of the equilibrium with 1 and 37, which are preferentially reduced.51 However, adding H2S to this equilibrium, in the absence of UV irradiation, converts cyanohydrin 44 to α-hydroxythioamide 43, which undergoes UV-promoted deoxygenation, providing thioamide 42. Prolonged irradiation reduces thioamide 42 further to an aldehyde which is trapped in the presence of HCN to form cyanohydrin 41. Ammonia converts 41 to valine Strecker intermediate aminonitrile 47. A further cycle of homologation by a similar addition/deoxygenation/reduction sequence furnishes the corresponding leucine precursor, aminonitrile 26 via α-hydroxythioamide 40, thioamide 39, and cyanohydrin 32.

Deoxygenation of α-hydroxyaldehydes, α-hydroxyketones, and α-hydroxythioamides prompted us to explore the same chemistry with other sugars of biological relevance. Interestingly, although their abiogenic provenance remains dubious, pentose sugars 48 (ribose or arabinose) can also be deoxygenated under the photoreduction conditions (likely via their open chain, aldehydic form), leading to deoxyribose 49, the carbohydrate component of DNA.50 Deoxygenation was observed for either isomer of 48 by irradiation with a catalytic amount of CuCN and H2S. As in the deoxygenation of 23 to 29, stoichiometric KCN was found to enhance the efficiency of the photoreduction, suggesting involvement in the catalytic cycle. Additionally, photoreduction of mercaptoethyl uracil 45 to thymine 46 was found to proceed in the presence of H2S, with or without a copper catalyst and thiocyanate. Considering the growing number of biologically implicated molecules we could synthesize via photoreduction and the related chemical network, it was apparent to us that UV irradiation was a critical driver of prebiotic chemistry.

Hence, we had demonstrated that the precursors of ribonucleotides, amino acids, and the hydrophilic component of biological membranes could have arisen simultaneously through a common chemistry, which is driven by UV light, uses H2S as a reductant, and can be mediated by Cu(I)–Cu(II) photoredox cycling. Although this chemistry compared very favorably with the formose reaction in terms of the yield and selectivity of its carbohydrate output, and provided a plethora of other biologically implicated molecules, the future of prebiotic chemistry lies in ever seeking to refine laboratory chemistry and the corresponding geochemical scenarios. The low solubility of H2S in water15 and relatively low abundance of copper in Earth’s crust34 would have restricted the cyanosalicidic network to specific and perhaps uncommon terrains, such as copper-rich and alkaline (pKa of H2S ≈ 7)35 pools. A more globally available reductant and more abundant metal as catalyst would suggest the photoreductive homologation chemistry of HCN may have been a ubiquitous primordial process. To these ends, we discovered that sulfite (SO32−) could fulfill the role of reductant and ferrocyanide could mediate the catalytic cycle, as well as provide a reservoir of cyanide.58 While both hydrogen sulfide and sulfur dioxide would have been pumped into the young Earth’s atmosphere by its highly active volcanic systems, and therefore both dissolved in terrestrial surface water, the various equilibria relating to Henry’s law, hydration, and dissociation lead to higher predicted concentrations of sulfite than hydrosulfide under most conditions.53 Sulfite, which also undergoes photoionization,49,50 is thus a more plausible ubiquitous source of hydrated electrons (and, depending on conditions, hydrogen atoms).

We found that irradiating (254 nm) a mixture of cyanide and bisulfite also promoted the photoreduction process, resulting in the production of bisulfite adducts of 14 and 15, 20 and 21, respectively (Scheme 2). Eventually, as bisulfite was removed from the equilibrium, homologation products 16, 19, and 24 were detected. Irradiation of a mixture of glycoaldehyde 23 and cyanide with bisulfite resulted in the formation of homologated (cyanohydrin of 31) and deoxygenated (29, 28, and 35) products. However, the reactions were not as efficient as the copper/hydrosulfide system. Ferrocyanide has been considered previously as a prebiotically plausible solution-phase repository of HCN 1 derived from the atmosphere, driven by the high association constant of Earth-abundant Fe2+ and cyanide.52 Ferrocyanide itself also provides hydrated electrons by photoionization,58 but we did not find it to be efficient as a sole reductant of cyanide or nitriles. We noted, though, that ferricyanide is known to be reduced by sulfite, forming ferrocyanide and sulfate.59,60 We therefore conceived a ferrocyanide/ferricyanide cycle with sulfite as a terminal reductant, attempted the photoreductive homologation with cyanide, sulfite, and catalytic ferrocyanide, and were delighted to find that it proceeded far more efficiently than the system with sulfite alone. Our experiments indicate that the photoredox cycle is mediated by ferrocyanide and its direct photooxidation, with sulfite serving as a two-electron reductant, either engaging directly as a terminal reductant, or first itself undergoing photooxidation to provide a hydrated electron, and the resultant sulfite radical monooxanion providing the second electron (Scheme 2 top, right). This system has the additional advantage of trapping aldehyde or imine products as their bisulfite adducts, 20, 21, 22, and 35, which are non-volatile and comparatively less reactive, plausibly allowing their potential concentration in evaporating pools of water. The equilibrium is displaced in favor of the free reactive aldehyde either as sulfite is removed from the reaction by its (photo)oxidation, or the addition of calcium cations and precipitation of calcium sulfite,61 allowing a prebiotic variant of the Knoevenagel–Bucherer modification of the Strecker reaction62 to proceed and furnish the network of biomolecules.

Systems chemistry can be a difficult balancing act—improvements to one system sometimes come with associated costs—for example, bisulfite, while improving the global plausibility of the photoreductive homologation chemistry of HCN, is known to catalyze the deamination of cytidine residues,63 and therefore may prove incompatible, in some way at least, with the chemistry providing RNA nucleosides. Incompatibility of functional groups, however, is by no means a problem rare in synthetic chemistry, and we continue to pursue reaction networks of increasing plausibility, compatibility, and consistency, by investigating the implications of one discovery on others.

4. PHOTOCHEMICAL NUCLEOSIDE SYNTHESIS

The nucleic acids RNA and DNA are among the most important molecules for the function of biology. A key tenet of any theory about the origin of known life is the availability of these or similar molecules, and therefore attempts at potentially prebiotic syntheses of genetic molecules, particularly of RNA, have been a focal point of prebiotic chemistry since its inception. While some approaches have yielded new insights into ways of assembling nucleosides and their

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derivatives, geochemically plausible and selective abiotic nucleoside synthesis is still an unfinished problem and an area of active research.

In 2009, our laboratory demonstrated that a selective synthesis of the RNA pyrimidine nucleotides from simple building blocks, such as C2 and C3 carbohydrates, was possible. This initial synthesis relied upon a photochemical conversion of cytidine-2',3'-cyclophosphate to a mixture of itself and uridine-2',3'-cyclophosphate. Aside from the photochemical provenance of carbohydrates discussed above, we have since discovered that nucleoside synthesis is remarkably enhanced by photochemistry, providing access to challenging reactivities and selectivities otherwise untapped by conventional thermal and catalytic means. That these transformations take place under conditions compatible with the geochemical scenarios hinted at by our carbon homologation chemistry (Section 3) seems like yet another clue (if only circumstantial) that our putative system might indeed overlap with the one(s) that led to the origin of life on Earth.

One of the key steps in our RNA pyrimidine synthesis was the union of C2 and C3 building blocks (the cyanamide adduct of 23 and 31, Scheme 2) to assemble aminooxazolines as a mixture of four diastereomers (ribo:arabino:lyxo:xylo = 44:30:13:8), which contain the pentose furanoside unit and are primed for nucleobase elaboration. The second-most prevalent isomer, arabino-30, with a β-configured anomeric

Scheme 3. Selective Prebiotic Synthesis of Pyrimidine Ribonucleosides and Purine Deoxyribonucleosides Driven by UV Irradiation

Non-canonical regioisomers are depicted in gray. Reactions are performed in water unless dry state or formamide (HCONH₂) is indicated. Ade = adenine, Pᵢ = inorganic phosphate.
Scheme 4. Proposed Mechanism of the Photoanomerization of 50/51

C–N bond, could be elaborated into the \( \beta \)-ribo-configured nucleotides, cytidine cyclophosphate and uridine cyclophosphate, via ultimate inversion at C2'.\(^5\) The fact that ribo-30 (RAO), with a non-canonical \( \alpha \)-configured C–N glycosidic bond, was the most prevalent aminoazoxaline diastereomer, was problematic, since neither RAO, \( \alpha \)-cytidine, nor \( \alpha \)-uridine can be efficiently isomerized to their 1'–\( \beta \)-isomers.\(^65\–67\)

Additionally, RAO spontaneously crystallizes when it forms, with enhanced enantiomeric excess if the initial mixture is non-racemic.\(^68\) These are exceptionally attractive characteristics of a synthetic intermediate in a likely cluttered and messy primordial soup, and the prospect that RAO might crystallize from a non-racemic soup which is then washed or drained away, leaving a chemically and enantiomerically pure starting point for RNA synthesis, prompted further evaluation of this intermediate.

Ultimately, combining the initial pyrimidine synthesis with our emerging, photochemical cyanosulfuric scenario, via the intermediacy of RAO (ribo-30), provided a much-improved prebiotic route to cytidine (52 C) and uridine (53 U) (Scheme 3). The discovery of a UV-induced photoisomerization between \( \alpha \)-2-thiouridine 50 and its anomer, \( \beta \)-2-thiouridine 51, was critical to this development.\(^70\) Reaction of RAO and cyanoacetylene S4 in aqueous phosphate buffer led to \( \alpha \)-anhydroriboctidine S5. Previously this compound had seemed a dead end because of its stereochemistry. In the context of the cyanosulfuric scenario, however, S5 undergoes efficient thiolysis by H2S, affording \( \alpha \)-2-thioribocytidine 50. After UV irradiation (254 nm), 50 is mostly anomerized (50:51 = 12.88) to provide \( \beta \)-2-thioribocytidine 51, a nucleoside of canonical stereochemistry. After hydrolysis of 51 in phosphate buffer or phosphorylation and hydrolysis, the canonical pyrimidine ribonucleosides (cytidine S2 C and uridine S3 U), or nucleotides thereof, are afforded in good yields, alongside 2-thioribonucleoside S8, a non-canonical nucleoside found in transfer RNA.\(^71\)

The mechanism of the photoanomerization between 50 and 51 was studied with deuteriation experiments and quantum chemical calculations, and was found to be analogous to the first step of a Norrish type II reaction (Scheme 4). Deuteration incorporation at Cl' was observed when \( \alpha \)-2-thiouridine 50 was irradiated in D2O, implying photoanomerization was associated with hydrogen atom abstraction at Cl', followed by inversion and/or deuteration. Calculations showed that \( \alpha \)-2-thiouridine 50 is readily photoexcited to a singlet state which, by virtue of the presence of its sulfur atom, undergoes facile intersystem crossing to the corresponding triplet state of \( \beta \)-\( \gamma \)-biradical 69. The proximity of Cl'–H and sulfur facilitates a rapid \( \beta \)-hydrogen atom transfer, giving 70, analogous to the \( \gamma \)-H transfer characteristic of type II Norrish reactions.\(^72\)

Once hydrogen atom abstraction occurs, the hydrogen atom, now attached to sulfur, becomes labile to hydrogen–deuterium exchange in D2O. Exchange is clearly rapid enough such that in some cases, it outcompetes anomerization of Cl', since deuteration of the alpha isomer was observed. Equilibrium ratios for 50:51 of 85:15 and 88:12 can be reached starting with 50 or 51, respectively. Interestingly, \( \alpha \)-2-thioribouridine does not undergo an analogous hydrogen atom abstraction, due to a greater distance between Cl'–H and sulfur, and the contrasting \( \pi \)-\( \pi \)-* and n–\( \pi \)-* characters of the triplet states for 50/51 vs uridine analogues. The strong preference for the \( \beta \)-isomer of 2-thiouridine at equilibrium provides a major pathway by which crystalline intermediate RAO can be transformed into the canonical pyrimidine ribonucleosides (cytidine S2 C and uridine S3 U). The propensity of the aminoazoxaline 30-forming reaction (Scheme 2) to favor the ribo-configured isomer was thus transformed from an Achilles’ heel of the previous synthesis into a defining strength of the current systems approach.

Introduction of sulfur into the nucleosides, via thiolysis of S5 to 50 (Scheme 3), enables the key photoanomerization (Scheme 4), but might seem like a diversion requiring the contrivance of additional thiolysis and hydrolysis steps. However, consistency with the cyanosulfuric scenario that produces carbohydrates and amino acids dictated this apparent diversion, and we were also drawn to the intermediacy of 2-thiopyrimidines, given their role in extant biology and their emerging importance in non-enzymatic replication of RNA studies conducted in the Szostak laboratory.\(^73\) Phosphorylation of cytidine S2 C and uridine S3 U provides oligomerizable\(^74\) cyclic phosphates of cytidine and uridine. This is an important step in the transition from a primordial soup of small molecules into macromolecules with biological function. Since 2-thioribonucleoside S8 is a co-product in the photoanomerization-based synthesis of pyrimidine ribonucleosides (Scheme 3), its phosphate is found in tRNA and it is a critical component in non-enzymatic RNA replication, we undertook to phosphorylate S8 under prebiotically plausible conditions (Scheme 3). We found that S’-phosphorylation of 2-thiouridine S8 occurred in hot formamide,\(^75\) providing 57, which should be able to undergo subsequent activation and oligomerization into primordial RNA sequences. Under complementary prebiotic phosphorylation conditions, though, in semi-molten urea,\(^76\) 2’-thiohydouridine 61 was produced (along with its phosphate esters). We recognized that this thiohydouridine, possessing a C2’–S bond, might undergo reduction to a 2’-deoxy derivative of RNA, which could serve as a building block for prebiotic DNA synthesis. Naturally, we gravitated toward the reduction conditions that we had already invoked in our photochemical cyanosulfuric scenario, and found that 61 could indeed be reduced in aqueous solution by the combined action of H2S, 2H2S, and D2O. Finally, we were able to isolate 2-thiouridine S8, the desired intermediate.
and UV light. After irradiation (254 nm) of 61 with H₂S for 3 h, non-canonical deoxynucleoside, 2′-deoxy-2-thiouridine 60 was afforded in 33% yield. The proposed mechanism (Scheme 5) of this photoreduction proceeds via photoionization of hydrosulphide, which provides a hydrated electron that reduces 61 to its radical anion, 71. 71 undergoes C₂−S homolysis to generate stabilized radical anion 72, which can abstract a hydrogen atom from H₂S to give the reduced product 60.

This generation of a deoxyribonucleoside intrigued us, as DNA has been widely considered to have arisen on Earth via the evolution of primitive ribonucleotide reductases or ribozymes, which convert ribonucleotides to deoxyribonucleotides in extant biology. However, although we demonstrated a prebiotic reaction reminiscent of the modern action of ribonucleotide reductases, we could only convert 60 to a canonical deoxynucleoside via a low-yielding dry-state transglycosylation. In this way, 60 served as a source of deoxyribose 49, by reaction with adenine 10 to provide deoxyadenosine 64a dA in 4% yield, or in solution by its facile hydrolysis to deoxyribose itself (hydrolysis half-time at 60 °C ≈ 30 h). While a prebiotic link between RNA and DNA had been revealed by this photoreduction, the low efficiency of the subsequent step in the synthesis of a canonical nucleoside was problematic, and even more so was the fact that the dry-state glycosylation provided a greater yield (6%) of the wrong stereoisomer of deoxyadenosine, α-64a (Scheme 3).

The successful conversion of pyrimidine thiophanhydrouridine 61 to a pyrimidine 2′-deoxyribonucleoside 60 prompted us to consider photoreduction of a purine thiophanhydrouridine 62, which might directly lead to the synthesis of a purine 2′-deoxyribonucleoside. Our strategy dictated that, in order to determine a prebiotic synthesis of such a molecule compatible with our previous findings, we return to our pyrimidine synthesis and attempt to find conditions under which any intermediates thereof might be diverted to thiohydropurine nucleosides. Here, our improved route via RAO and subsequent intermediates 55 and 50 then 56 proved key: thiohydropurin deoxynucleoside 62 is formed in high yield by the tethered glycosylation of α-2,2′-anhydrocytidine 55 or α-2,2′-anhydrothiouridine 56 with glycosyl acceptor nucleophile 8-mercaptopurine 59, upon heating in the dry state with MgCl₂ (Scheme 3). 59 itself is available by dry-state combination of adenine hydrolysis product 4,5,6-triaminopyrimidine and thiocyanate, itself a product of our cyanosulfidic photoreduction chemistry. Unfortunately, we had another dry-state selectivity problem: both canonical N⁷-regiosomer 62 and its non-canonical N⁷-regiosomer, 63, were products of the reaction, with the latter the dominant species in a 45:55 ratio. Hydrosulphide photoreduction of 62 and 63 would lead to C₂′-reduced nucleosides 67a and 68, respectively (Scheme 3 and Scheme 6a–c), which, via a mechanism recently reported by Powner et al. (Scheme 6d), can undergo further photoreduction to deoxyadenosine 64a dA, and its non-canonical N⁷-regiosomer 66 (Scheme 3). The problem of the selectivity of the dry-state tethered glycosylation was solved unexpectedly by the discovery that the photoreduction of the purine thiophanhydrouridine precursors proceeded with canonical selectivity (Scheme 4). When H₂S was used as the reductant, a 45:55 mixture of 62 and 63 led to a photoreduced mixture of 64a dA and 66 (60:40 ratio). This canonical selectivity could be further enhanced by the relative hydrolysis rates of the two products: the non-canonical N⁷-regiosomer of deoxyadenosine (66) was found to be 70 times more labile to hydrolysis than its relatively stable canonical counterpart (64a). Remarkably, when sulfitelbisulfitel was used as the reducing agent, only the N⁹-regiosomers of purine deoxyribonucleosides 67a and 64a dA could be detected after photoreduction. The non-canonical N⁷-isomer 63 in the mixture was photochemically destroyed, which we verified by performing the photoreduction on pure 63.

Quantum chemical calculations combined with experiment revealed the origins of this selectivity and explained the different outcomes for the two reducing agents (Scheme 6). In the sulfitelbisulfitel system, two competing possible mechanisms were proposed (Scheme 6a,b). In the first (Scheme 6a), photolysis of 62 was calculated to favor C₂′−S homolysis (73) via the lowest excited singlet state, which in the presence of reducing agents provides 67a and ultimately 64a dA. In contrast, photooxidation of 63 leads to C₈−N₇ homolysis (74) and destruction of the material. Alternatively, direct reduction of the canonical N⁹-thiohydropurin 62 by a hydrated electron (photodetached from sulfitelbisulfitel) was shown to favor C₂′−S bond cleavage to produce radical anion 75 (Scheme 6b), analogous to our previously proposed mechanism for the reduction of 61 (Scheme 5). However, the C₂′−S bond of 63 was not found to be susceptible to reduction, with 63 instead undergoing C≡N π-bond cleavage to 76 and likely subsequent decomposition. In the case of hydrosulphide as the terminal reductant (Scheme 6c), although a similar reduction pathway proceeding via a photochemically generated electron is possible, stable encounter complexes of the thiohydropurines and hydrosulphide featuring chalcogenic electrostatic S···S interactions were located, explaining the observation of both N⁹ and N⁷ reduced products. These encounter complexes (including 77) both favor C₂′−S bond cleavage via a UV-induced charge transfer (CT) from hydrosulphide to the thiohydropurine resulting in reduction products for both 62 and 63, via radical anions 75 and its N⁷ regiosomer (Scheme 6c). 67a and the corresponding N⁷ regiosomer 68 is presumably desulfurized via the mechanism described for similar 8-mercaptopurine nucleosides by Powner et al. (Scheme 6d). As well as its clear relevance to the prebiotic reduction of thionucleosides, this...
Scheme 6. Proposed Mechanism of Photoreduction for Thioanhydroadenosines with Bisulfite or Hydrosulfide, and C8–S Photoreduction of the Products Generated and Related Nucleosides

“Photoreduction with bisulfite may proceed via competing mechanisms in (a) and (b). In (a), photolysis of the thioanhydronucleoside occurs before reduction, providing different intermediates in the reduction of N9-thioanhydroadenosine 62 and N7-thioanhydroadenosine 63, and thus different outcomes. In (b), reduction of 62 and 63 is effected by a photochemically generated hydrated electron, resulting in different radical anion intermediates and different reaction outcomes. (c) Stable encounter complexes of the thioanhydroadenosines and hydrosulfide such as 77 were calculated, and allow the reduction to proceed for regioisomers 62 and 63 in a similar fashion, with radical anion 75 and its N7-regioisomer as intermediates (mechanism for 63 not shown). (d) Photoreduction (300 nm) of 67a to generate 64a presumably proceeds via the mechanism calculated by Powner et al. for related ribo- and arabino-8-mercaptopurine nucleosides 67b–g. The yield of ribo- and ara-inosine (64f and 64g) was low because of a competitive photodecomposition pathway via 80. CT = charge transfer.

novel protective photoreduction effect that hydrosulfide exerts may be of broader relevance in photochemistry.

Interestingly, the photochemical desulfurization of 8-mercaptopurine nucleosides 67, of either ribo or arabino configuration, was also found by Powner et al. to be inherently selective.82 Ribo- and arabino-adenosine (ribo-A 64b, ara-A 64c) and ribo- and arabino-guanosine (ribo-G 64d and ara-G 64e) were produced in high yield from their respective 8-
mercaptopurine nucleoside starting materials (67d and 67e) by irradiation (300 nm), whereas ribo- and arabino-inosine (ribo-I 64f, ara-I 64g) were produced in much lower yields. Experiment, theory, and femtosecond scale spectroscopy identified photolysis of the C=5 π bond to generate redox-active triplet species 78, which extrudes triplet sulfur to form N-heterocyclic carbene 79, and tautomerization to the corresponding reduced nucleoside (ribo- and ara-A 64a and 64c), G (64d and 64e), and I (64f and 64g) as the productive mechanism. For the case of the inosine derivatives, an alternative, long-lived triplet state 80 was found to be more stable than the competing equilibrium redox-active triplet state and resulted in decomposition of material by bimolecular radical reactions. Selection based on photostability or photochemical mechanisms is thus a property of nucleosides and their synthesis that seems too effective to ignore in origins of life scenarios.84,85

Deoxyadenosine 64a is (partially) converted slowly via hydrolysis or more rapidly via nitrosative deamination to deoxyinosine 65 dI (Scheme 3). Ultimately, therefore, our diversion via RAO (ribo-30) had led to a prebiotically plausible synthesis of pyrimidine ribonucleosides cytidine (C) and uridine (U), and purine deoxyribonucleosides deoxyadenosine (A) and deoxyxinosine (dI), via respective routes diverging from 50 or 55. The route to the pyrimidines featured a critical photoanomerization, and the route to the purines a critical photoreduction, with hydrogen sulfide and/or hydrogen sulfite being intimately associated with these steps, as well as the synthesis of the carbohydrate precursors of RAO itself.

5. CONDENSATION CHEMISTRY ENABLED BY PHOTOCHEMISTRY

One of the key challenges in mapping a transition between chemical building blocks and primordial biological systems is the condensation of oligomers into functional polymeric derivatives. Condensation of phosphates and alcohols, carboxylic acids and amines, and carboxylic acids and alcohols leads to nucleic acid polymers, polypeptides, and peptidyl-RNA, respectively (Scheme 7). In aqueous solution these reactions are thermodynamically disfavored, and therefore their occurrence requires the system to be disturbed from equilibrium, usually by the delivery of reactive, so-called activating or condensing reagents, which mediate the dehydration conjoining reactions described above. Methyl isonitrile 81 is one such molecule, as it facilitates condensation reactions driven by the formation of σ-bonds at the expense of the π-bond components of its high chemical potential triple bond, resulting in a dehydration reaction and formation of the hydrated byproduct amide 82. Our laboratory investigated the photochemically mediated synthesis, and activation chemistry, of methyl isonitrile 81. We discovered that mixtures of simple and stable reagents such as ferrocyanide, nitrite salts, and simple amines such as methylamine, under the action of UV irradiation, are able to provide a source of methyl isonitrile 81 (Scheme 8).86

Scheme 8. Prebiotically Plausible Photochemical Synthesis of Methyl Isonitrile 81

In summary, ferrocyanide 83 undergoes photoaquation to form transient complex 85, which undergoes ligand substitution of water with nitrite to ultimately form nitroprusside 87. Nitroprusside 87 reacts with methylamine 89 to form alkyating species 88, reaction of which with ferrocyanide forms isonitrile complex 86. Irradiation of 86 in the presence of a displacing ligand such as cyanide provides methyl isonitrile 81.

The cycle producing methyl isonitrile 81, elucidated by experiment, is depicted in Scheme 8. Ferrocyanide 83, under UV irradiation, undergoes photoaquation to transiently form [Fe(CN)3H2O]3− 85, which in the presence of nitrite is converted to diazotizing agent nitroprusside 87. This conversion is fastest at relatively long wavelength UV irradiation (360 nm), but we also demonstrated that it occurs at 254 nm. Reaction of nitroprusside 87 and methylamine 89 generates stabilized iron(II) diazonium complex 88, which
alkylates ferrocyanide $83$ to generate iron(II) isonitrile complex $86$ and liberates $84$, from which nitrogen gas dissociates and may be replaced by cyanide, regenerating ferrocyanide $83$. Nitrogen can re-enter the cycle after being oxidized in the (CO$_2$-rich) atmosphere by electrical storms or shock waves, and dissolution and disproportionation in water, as nitrite.$^{87}$ Complex $86$ can accumulate until release of methyl isonitrile $81$ is triggered by UV irradiation, releasing an activating agent into the surrounding aqueous environment. Association of cyanide replaces the departing isonitrile and regenerates ferrocyanide $83$. It seemed possible, therefore, that day-night cycling could lead to different periods of UV-promoted synthesis of nitroprusside $87$, followed by accumulation of $86$ in the dark with inflow of methylamine $89$, followed again by a UV-promoted day cycle of methyl isonitrile $81$ release. Indeed, irradiation of a mixture derived from ferrocyanide $83$, nitroprusside $87$, and methylamine $89$ showed production of methyl isonitrile $81$ in 13% yield. Although possible in a prebiotic context, this synthesis path is not yet as robust as some of the other chemistry we have

Scheme 9. Various Methods of Activating Phosphate and Carboxylate Groups$^a$

(a) The Passerini reaction is an efficient method to activate phosphate monoesters; however, carboxylates rearrange intramolecularly once activated. (b) Methyl isonitrile $81$ in combination with DCI $101$ facilitates both phosphate and carboxylate group activation; thus, upon activation at moderately low pH, a mixture of nucleoside phosphates and (acyl)amino acids provided peptides, mixed anhydrides, and aminocacyl RNA. (c) The DCI/methylisonitrile-mediated condensation activates amino acids to form racemizing intermediates $110$, which, for acetylalanine $104$, are partially resolved by A$3^\prime$P. AMP = adenosine 5$'$-monophosphate. Ade = N9-adenyl.
discovered, and further work is continuing to refine it. We moved ahead to study the activation chemistry enabled by isonitriles such as 81 because we felt it would tell us about the requirements for prebiotic activation chemistry in general.

Initially, the condensing power of isonitriles was utilized via a prebiotic variant of the Passerini\textsuperscript{98} multicomponent reaction (Scheme 9a).\textsuperscript{94,95} At near neutral conditions, addition of methyl isonitrile 81 to an aldehyde such as acetaldehyde 90 provides a highly electrophilic cationic nitrium ion 91 (or cyclized imino-\(\alpha\)-lactone thereof), which can undergo addition from a condensation partner such as phosphate. The newly formed imidoyl phosphate intermediate 92 is an activated phosphate which can be trapped by imidazole 93, to form phosphoroimidazolides 95. A similar reaction, analogous to the Ugi multicomponent reaction,\textsuperscript{91} can proceed using imines in place of acetaldehyde.\textsuperscript{90} Phosphoroimidazolides such as 95 are the necessary activated nucleoside phosphates in the most efficient prebiotic oligomerization and non-enzymatic replication chemistries.\textsuperscript{85} At pH 6.5, the reaction of a nucleoside S’-phosphate (AMP, GMP, UMP, or CMP), acetaldehyde 90, and methylisonitrile 81 gave up to 75% yield of phosphorimidazolide 95. Additionally, multiple separate additions of condensing agent methyl isonitrile were shown to continuously activate phosphate groups after repeated cycles of hydrolysis, apparently uninhibited by the formation of hydrolysis byproducts such as 94. Another advantage of this prebiotic condensation chemistry was its selectivity for phosphate; no modification of nucleobases was detected in any of the reactions. However, the Passerini (and Ugi) reaction mechanism limited the combination of aldehydes and methyl isonitrile to the potential oligomerization of phosphate groups; carboxylic acids 96 (including amino acids) react in an intramolecular fashion, ultimately providing esters 98 via acyl transfer of intermediate 97. (Scheme 9a).

We ultimately discovered that methyl isonitrile activation chemistry could be extended to encompass both phosphate and carboxylic acid groups, by replacement of the aldehyde in the Passerini reaction with the simplest of electrophiles, a proton, and a prebiotically plausible nucleophilic catalyst, 4,5-dicynoimidazole (DCI) 101 (Scheme 9b,c).\textsuperscript{95–98} At a pH 4–5.2 (the p\textsubscript{K\textsubscript{a}} of DCI), methyl isonitrile can undergo addition by N-protected amino acids and carboxylic acids, and more slowly by phosphate. In the presence of DCI (and absence of aldehydes), activated imidoyl-carboxylates 97 react with S’- and 3’-nucleotides to provide activated nucleotides (2’,3’-cyclic AMP, cAMP 102), mixed phosphate-carboxylate anhydrides (e.g., 106, 107), and 2’-(acyl/peptidyl) nucleotides (e.g., 108, 109); with themselves to provide protected peptides (e.g., 103); and with glycerol-2-phosphate to form lipidated glycerol-2-(cyclic)phosphate esters. Aside from working on isolated samples, an example of mixed reactivity is shown in Scheme 9b. As such, DCI acts to enhance the reactivity of activated phosphates and carboxylates, in an analogous manner to its use as an efficient activator in phosphoramidite-based oligonucleotide synthesis.\textsuperscript{96} Separately, a slight kinetic preference for the ligation of protected L-alanine (vs protected D-alanine) with model D-ribonucleotides was also identified, and (limited) dynamic kinetic resolution in aminoacylation, via a racemization pathway featuring 111, was observed: processes which might have played a role in the development of the preference seen in extant biology for D-carbohydrates and L-amin acids (Scheme 9c). The conditions, in combination with divalent metal cations such as Mn\textsuperscript{2+}, also affected the ligation of RNA oligomers in up to 72% yield at pH 6. Cyclization of A3’P 99 to cAMP 102 could also be performed after in situ UV-mediated (360 nm) release of methyl isonitrile 81 from its iron complex 86. Thus, photochemically generated methyl isonitrile 81 may have been a versatile prebiotic activating agent.

6. PHOTOCHEMICAL STABILITY AND PHOTOSELECTION

UV radiation is a double-edged sword with respect to chemical synthesis. As described above, UV radiation provides energetic input to form bonds and selectively synthesize important biomolecules and their precursors. In many conditions, though, UV radiation will lead to the breakdown of such organic matter, via photolysis. The danger that UV light presents to modern biology, especially genetically encoded information, is evident in the evolution of various mechanisms to protect organisms from it.\textsuperscript{85} Before these mechanisms developed, the abiological chemical synthesis of biological building blocks would have had to benefit from more primitive mechanisms. These could have been provided by the environment, and could have been highly variable, potentially allowing a favorable balance between UV-promoted photochemistry and equally important processes occurring in the dark. Such mechanisms include day/night cycling, climate, water depth and turbidity, terrestrial features, rock pores, mineral surfaces, and the presence of UV-absorbent sunscreen molecules in mixtures.\textsuperscript{97,98} The canonical nucleosides enjoy remarkably high photochemical stability relative to non-canonical derivatives, stemming from ultrafast non-radiative photochemical relaxation pathways, which effectively allow UV energy input to be successfully dissipated as heat rather than chemical modification.\textsuperscript{85} Additionally, simple nucleic acid polymers may benefit from innate light-mediated repair mechanisms for photoinduced damage. Thus, prebiotic chemistry may not have had to rely long on environmental factors before native physical and chemical protection mechanisms allowed mixtures

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**Scheme 10. Photochemical Input Will Act to Synthesise and Destroy Molecules—The Chemical Evolution of Prebiotic Mixtures, Based on Mixture-Variable Rates of Synthesis and Destruction, Could Present an Explanation for the Preponderance in Biology of Typically Photostable, Canonical Biomolecules**

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Scheme 11. An Example of Photochemical Stability Placing a Constraint on the Prebiotic Scenario

In this case, the photochemical stability of 113 is low compared to its forward reaction rate in the sequence, thereby imposing concentration minima or sunscreens on the mechanism.

sufficient resistance to UV radiation to benefit from photochemical synthesis and selection while resisting photochemical degradation (Scheme 10).

The highly resistant photophysical and -chemical properties of nucleoside monomers and polymers are consistent with having emerged from an environment with UV-resistance as a selected-for characteristic. Extrapolating the presence of UV radiation to the earliest stages of prebiotic chemistry provides a basis for exploring the photochemical synthesis described above, but also presents an additional potential physical constraint with which to evaluate it. Thus, the quantity of UV flux and its wavelength distribution at the surface of early Earth, in potentially different geochemical environments, need to be modeled and applied to newly discovered photochemistry in a quantitative way where possible. This includes the evaluation of the photochemical stabilities of prebiotically interesting intermediates (and crucially, mixtures) and an investigation of the UV-shielding mechanisms likely to have affected them. Encouragingly, quantitative and kinetic studies modeling plausibility of recently proposed chemistry and the photochemical stability of some important intermediates are emerging.3,100−104 An important illustration of the future direction of these studies is the series of work by Todd et al., who evaluated the photostability of three prebiotically interesting heterocycles, including 2-aminooxazole (113, Scheme 11), an intermediate in the synthesis of ribo-30 (RAO) (Scheme 2, 23 + 25 → 113).102 These studies revealed the wavelength dependence of the non-specific photo-degradation of 2-aminooxazole 113, which was 3−10 times faster below 255 nm than above it. Assessing its stability over the whole spectrum likely available to a prebiotic Earth yielded a half-life of 7 h for 2-aminooxazole 113. The authors concluded that under these model irradiative conditions, the unbuffered reaction between glyceraldehyde 31 and 2-aminooxazole 113 (to form the more photochemically robust103 aminooxazolines 30, Schemes 2 and 11) would need proceed at rates only occurring when the concentration of both substrates is greater than 0.1 mM to exceed the rate of the photodecomposition of 113 (measured in isolation). This is an encouraging start toward placing reasonable kinetic constraints on parts of sequences invoking photochemical steps, that may necessitate specific sunscreen mechanisms. As part of the process of refinement described throughout this Perspective, invoking a specific sunscreen mechanism could have interesting consequences on the chemistry in question. Todd et al. have since described further progress toward such a constraint, where they show that 113 is increasingly stable at higher concentrations, and that simple model prebiotic mixtures containing nucleosides provide modest sunscreen effects (e.g., a 3-fold increase in the persistence of 2-aminooxazole 113 in the presence of 0.1 mM adenosine).104 Clearly, further work toward characterizing the (photo)-stabilities of prebiotic mixtures is required in this specific, and a broader, context. Finally, we note here that photochemical stability considerations are applicable to all prebiotic chemistries that may have occurred on Earth's surface, not just those that explicitly invoke photochemical transformations. This emphasizes the importance of characterizing potential sunscreens and how they may affect the chemistry being evaluated.

It is difficult to predict the outcome of photochemical sequences, particularly with respect to relative photodegradation rates, based on isolated studies, since these rates seem highly dependent on the chemical environment, and can be affected by photosensitization and chemiluminescence, factors which in this context are yet to receive significant experimental attention. Such isolated studies are still important, as deviation from their predictions implies interesting chemical phenomena at play. While the complexity of such studies in the context of potentially prebiotically interesting mixtures is daunting from an analytical point of view, embracing this challenge with increasingly sophisticated experimental strategies and analytical techniques, with an ever growing body of relevant kinetic and physical data, is an important step forward for origins-related chemical research.

7. CONCLUSION AND FUTURE PERSPECTIVE

Albert Eschenmoser used the term “chemomimetic” to describe those few biosynthetic reactions which clearly map onto inherently favored, non-enzymatic chemical reactivity of the substrates.105 As chemists humbled by the way that biology juggles thousands of different reactions in water at close to room temperature, Eschenmoser’s words are good to hear. However, the majority of primary metabolic reactions are not “chemomimetic”. They are not inherently favored and require highly efficient enzyme catalysis to bring their reaction rates into a realm that makes them biochemically useful. Indeed, were many such reactions to be efficient in the absence of catalysis, metabolic regulation would not be possible—there would be no off switches. Biology does not invent new reactions, to use Jeremy Knowles’s wonderful turn of phrase, but it can clearly be seen to have selected reactions that are slow in the absence of catalysis. This implies that these reactions were first deployed by biology when rudimentary catalysts were already available. For a few reactions, these catalysts could have been metal ions, surfaces, and so on, but...
for the majority, ribozymes or enzymes would have been necessary. It is thus likely that a large part of primary metabolism was only acquired after RNA sequence space had started to be explored, and maybe only after the advent of coded peptide synthesis. Evolution over the aeons has then provided the incredibly efficient (predominantly) enzyme catalysts that we see today. But, to get to the point that RNA and coded peptides were being explored for catalytic potential, biology must have either synthesized its building blocks using chemistry different to that underlying extant metabolism, or had them provided by the environment. The former case would require efficient synthesis of four nucleotides, lipids, and, say, 10 amino acids in one pot (a cell), with minimal biocatalysis, under biocompatible conditions, and the chance of this being possible strikes us as vanishingly small. In the latter case, the chemistry must again operate without sophisticated biocatalysis, but now it does not have to be constrained by being one pot and biocompatible.

Prebiotic routes to the key nucleotide, lipid, and amino acid products must have been inherently favored, however, to avoid systems degenerating into overly complex mixtures and for synthesis to outcompete degradation. Highly complex mixtures are problematic because of the dilution of individual components and the fact that conversion to oligomeric structures then compounds the complexity. How could nascent biology deal with having to find the things it needed amidst a molecular mess when it was surely confronted by many, many other problems? Surely it would be easier for life to develop if it was just provided by the environment with the things it needed and not much else besides? So, for most compounds and simple systems, it is perhaps best to look for highly efficient synthetic prebiotic chemistry that is different to biochemistry, can be bioincompatible, and can involve some degree of spatial or temporal separation. Given such chemistry and subsequent mixing by, for example, fluvial action, stockpiles of key compounds could have been accumulated and mixed over long periods to provision the origin and early evolution of life when conditions became conducive.

How then can we say whether a potentially prebiotic reaction network discovered in the laboratory actually operated on early Earth? There can be no certainty, but we would say that is most likely when a prebiotic reaction network makes clear the synthetic links between biomolecules that do not appear connected when seen from a purely structural point of view, for then a logical chemical explanation for the nature of biological componentry is provided. The (sequences of) conditions for the most compelling syntheses and networks should then be taken as clues to develop geochemical scenarios that would support and facilitate the chemistry. Other consequences of these scenarios can then be fed back into the synthetic chemistry, and if the latter responds favorably by producing other biomolecules or giving fewer undesirable byproducts, both the chemistry and the scenario become more likely representative of what actually took place on early Earth. Key tenets of future research will therefore be continual refinement of plausibility and an increasing embrace of the complexity of systems chemistry.

For example, the diverse photochemistry of building blocks such as cyanide and its derivatives stimulates the question of whether such building blocks could become available under the same photochemical conditions they are then subjected to, or what kind of geochemical scenarios might reasonably lead to their stable accumulation. Provision of atmospheric hydrogen cyanide, for example, has been discussed in a variety of atmospheric contexts, with its most likely source being the delivery of reduced material (for example metallic iron) via impactors, which in weakly reducing CO₂/N₂ atmospheres can lead to thermal and photochemical production of hydrogen cyanide. The transient reducing power of the impactor favors the production of organics, which has also been postulated to lead to UV-attenuating hazes. Thus, if hazes did accompany hydrogen cyanide production, for cyanide to undergo surface photochemistry, it must have accumulated and remained on the surface until the haze dissipated. Such considerations have led to the proposal of alkaline carbonate lakes as stable reservoirs for cyanide (as sodium ferrocyanide and/or ferrous ferrocyanide) over a wide range of temperatures and CO₂/HCN atmospheric compositions, where ferrocyanides could accumulate and persist beyond any haze, providing a reservoir of sodium cyanide via eventual thermolysis. Thus, exploration of the chemistry prompts geochemical considerations, providing focus on prebiotic environments of greatest interest. As a corollary, the carbonate lake setting precludes the formation of calcium ferrocyanide, which has been invoked as the precursor (by thermolysis) to cyanamide (Scheme 2) in prebiotic nucleoside syntheses. This problem spurred the discovery of an alternative, sodium ferrocyanide-mediated photochemical synthesis of cyanamide. This provides an example of exchange of information between what is feasible chemically and what is implied geochemically, a continuing process of refinement that generates prebiotic chemical hypotheses of increasing plausibility and consistency.

It may seem counterintuitive that we should embrace selective chemistry and systems chemistry at the same time. After all, synthetic chemistry is traditionally viewed and practiced as the conversion of A to B by reaction with C, for ease of preparation and analysis. Conventionally, selectivity is viewed as high if it is for a single product, but this need not be the case, and we need to remove these traditional limits on how many materials we start and finish with in a chemical reaction. This does not mean we should be satisfied with chemistry generating complex and dilute mixtures of biological and related molecules (the Murchison meteorite contains several million compounds, but, rather, that we look for predisposition toward not just one target, but many useful compounds, while avoiding undesired mess. In a related sense, chemical yield is less important in a prebiotic context than it is in traditional synthesis, and it is the composition of the final mixture of products that is important. While having high yields in key sequences of reactions is one way of achieving this, it is not the only one, as purification of an intermediate resets the composition. Of the various purification methods known in conventional chemistry, crystallization seems highly plausible in an early Earth environment. Separation of crystals from the mother liquor could perhaps be most easily achieved through fluvial action: a dwindling, slow-moving stream saturated in a compound, depositing crystals that subsequently dissolve following rainfall, with the stream then following a different path. Indeed, even an initially low yielding process can be of extreme prebiotic interest if a key intermediate crystallizes from solution. When the crystals dissolve in a fresh solution and new chemistry ensues, giving a broad palette of (proto)biomolecules, the low yield of the initial process is inconsequential given sufficient material flux. Systems (photo)-
chemistry in an origins of life context is an immature area, but an extremely exciting one as our ability to analyze and understand the very complexity necessary for life progresses quickly.\textsuperscript{5,110} Exploring chemical space is easier via systems chemistry, and indeed the very nature of life itself precludes the traditional synthetic approach.

The utility of UV light in wide-ranging prebiotic processes that satisfy the foregoing criteria has important implications for the environmental constraints on the origin of life, which warrant further investigation. First, the importance of photochemistry helps us hone geochemical scenarios by elimination, and clearly points away from the experimentally unsupported, yet somehow still advocated, deep sea hydrothermal vent origin of life theories.\textsuperscript{51} Invoking UV-mediated photochemical synthesis also necessitates reasonable mechanisms by which a prebiotic inventory of increasing complexity, especially any particularly photosensitive intermediates, can be protected from constant or excessive UV irradiation. Synthesis of organic matter in a photosynthetic, but also potentially photodestructive environment allows the prediction of kinetic and timeline constraints on the chemistry, and where special mechanisms for increased photochemical persistence are invoked, their effects on the systems chemistry must be evaluated. Thus, such constraints can provoke the discovery of new chemistry. Understanding exactly what balance of these mechanisms is required requires more quantitative studies, of increasing sophistication.

We also suggest that selection taking place via photochemical processes should be further explored. In our own work we have found chemical processes often exhibit remarkable, unpredictable selectivity in favor of molecules that are utilized by extant biology. Two instructive examples are the selectivity in the photoepimerization of \textit{SO} (Scheme 4)\textsuperscript{50} and the selective photodestruction of the non-canonical \textit{N}-isomer of thioanhydroadenosine, \textit{63} (Scheme 3).\textsuperscript{51} Selectivity such as that observed in these reactions, can be considered as a circumstantial implication that such processes may have contributed to the origins of life, in contrast to unselective processes that result in the properties and functions of biomolecules being lost in a myriad of contaminants. In a systems level context, if a key environmental input such as UV radiation is implicated in this way, a rigorous evaluation of the viability of other molecules and processes in this environment becomes prudent, to evaluate their joint plausibility. Instability of one set of molecules in the conditions necessary for the formation of others does not necessarily render them mutually exclusive in a broad origins of life context, but certainly points to the need for the development of variants of such a scenario and helps us evaluate general plausibility. This is epitomized in the culmination of our prebiotic (deoxy)ribonucleoside synthesis work (Scheme 3).\textsuperscript{51} A critical element of this work was the separate evaluation of the prebiotic synthesis of purine deoxynucleosides and pyrimidine ribonucleosides, and then a systems level evaluation of their joint plausibility. The latter revealed that a synthesis of set of four nucleosides is indeed possible under the same chemical conditions. A remarkable subtlety of this system that only emerges at this level is the fact that the pyrimidine nucleosides \textit{C} \textit{52} and \textit{U} \textit{53} are not stable to the photoreductive synthesis of the purine deoxynucleosides \textit{dA} \textit{64a} and \textit{dl} \textit{65} in isolated circumstances, but, in the presence of the purine deoxynucleoside intermediates \textit{62} and \textit{63}, they persist, and a synthesis of all four nucleosides is thus only possible as a mixture. Such systems level evaluation needs to become a benchmark for plausibility in prebiotic synthesis.

A striking aspect of the progress that has been made in prebiotic chemistry since its early days is the shift away from guessed-at geochemical scenarios that turn out to only support unselective and low-yielding chemistry, toward both chemically and geochemically inspired scenarios that are high yielding and/or selective, by virtue of chemical selectivity or geophysically plausible purification mechanisms. Allowing the chemistry to guide our refinement of geochemical scenarios enables us to focus on those that are most probable to have formed mixtures rich in (proto)biomolecules and, crucially, little else. A dialogue between chemists and our colleagues in earth and planetary sciences and astronomy is crucial to advance this venture—only together will we solve the origin of life conundrum.

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\section*{Notes}

The authors declare no competing financial interest.

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\section*{REFERENCES}

\begin{enumerate}
\item Yadav, M.; Kumar, R.; Krishnamurthy, R. Chemistry of Abiotic Nucleotide Synthesis. \textit{Chem. Rev.} \textbf{2020}, \textit{120} (11), 4766–4805.
\item Sutherland, J. D. The Origin of Life - Out of the Blue. \textit{Angew. Chem., Int. Ed.} \textbf{2016}, \textit{55} (1), 104–121.
\item Sutherland, J. D. Opinion: Studies on the Origin of Life — the End of the Beginning. \textit{Nat. Rev. Chem.} \textbf{2017}, \textit{1} (2), 0012.
\item Sasselov, D. D.; Grotzinger, J. P.; Sutherland, J. D. The Origin of Life as a Planetary Phenomenon. \textit{Sci. Adv.} \textbf{2020}, \textit{6} (6), No. eaax3419.
\item Ruiz-Mirazo, K.; Briones, C.; de la Escosura, A. Prebiotic Systems Chemistry: New Perspectives for the Origins of Life. \textit{Chem. Rev.} \textbf{2014}, \textit{114} (1), 285–366.
\item Cnosken, I.; Sanz-Forcada, J.; Favata, F.; Wittase, O.; Zegers, T.; Arnold, N. F. Habitat of Early Life: Solar X-Ray and UV Radiation at Earth’s Surface 4–3.5 Billion Years Ago. \textit{J. Geophys. Res.} \textbf{2007}, \textit{112} (E2), E02008.
\item Hulet, H. R. Limitations on Prebiological Synthesis. \textit{J. Theor. Biol.} \textbf{1969}, \textit{24} (1), 56–72.
\item Miller, S. L.; Urey, H. C.; Oró, J. Origin of Organic Compounds on the Primitive Earth and in Meteorites. \textit{J. Mol. Evol.} \textbf{1976}, \textit{9} (1), 59–72.
\item Miller, S. L. A Production of Amino Acids Under Possible Primitive Earth Conditions. \textit{Science} \textbf{1953}, \textit{117} (3046), 528–529.
\end{enumerate}
isomerization: An Intermediate Step in the Prebiotic Formation of Purine Nucleobases. Chem. - Eur. J. 2014, 20 (9), 2515–2521.
(33) Ferus, M.; Kubelik, P.; Knizek, A.; Pastorek, A.; Sutherland, J.; Civis, S. High Energy Radical Chemistry Formation of HCN-Rich Atmospheres on Early Earth. Sci. Rep. 2017, 7 (1), 6725.
(34) Miyakawa, S.; James Cleaves, H.; Miller, S. L. The Cold Origin of Life: A. Implications Based on the Hydrolytic Stabilities of Hydrogen Cyanide and Formamide. Origins Life Evol. Biospheres 2002, 32 (3), 195–208.
(35) Smith, K. E.; House, C. H.; Arevajo, R. D.; Dworkin, J. P.; Callahan, M. P. Organometallic Compounds as Carriers of Extraterrestrial Cyanide in Primitive Meteorites. Nat. Commun. 2019, 10 (1), 2777.
(36) Ritson, D.; Sutherland, J. D. Prebiotic Synthesis of Simple Sugars by Photoredox Systems Chemistry. Nat. Chem. 2012, 4 (11), 895–899.
(37) Ritson, D. J.; Sutherland, J. D. Synthesis of Aldehydic Ribonucleotide and Amino Acid Precursors by Photoredox Chemistry. Angew. Chem., Int. Ed. 2013, 52 (22), 5845–5847.
(38) Xu, J.; Ritson, D. J.; Ranjan, S.; Todd, Z. R.; Sasselow, D. D.; Sutherland, J. D. Photochemical Reductive Homologation of Hydrogen Cyanide Using Sulfite and Ferrocyanide. Chem. Commun. 2018, 54 (4), 5566–5569.
(39) Ritson, D. J.; Sutherland, J. D. Conversion of Biosynthetic Precursors of RNA to Those of DNA by Photoredox Chemistry. J. Mol. Evol. 2014, 78 (5), 245–250.
(40) Patel, B. H.; Percivalle, C.; Ritson, D. J.; Duffy, C. D.; Sutherland, J. D. Common Origins of RNA, Protein and Lipid Precursors in a Cyanosulfidic Protometabolism. Nat. Chem. 2015, 7 (4), 301–307.
(41) Horváth, A.; Papp, S.; Déczy, Z. Formation of Aquated Electrons and the Individual Quantum Yields for Photoactive Species in the Cu(I)-KCN-H2O System. J. Photochem. 1984, 24 (4), 331–339.
(42) Katagiri, A.; Yoshimura, S.; Yoshizawa, S. Formation Constant of the Tetracyanocuprate(I) Ion and the Mechanism of Its Decomposition. Inorg. Chem. 1981, 20 (12), 4143–4147.
(43) Kilián, H. Ueber Das Cyanhydrin Der Lävulose. Ber. Dtsch. Chem. Ges. 1885, 18 (2), 3066–3072.
(44) Fischer, E. Reduction von Säuren Der Zuckergruppe. Ber. Dtsch. Chem. Ges. 1889, 22 (2), 2204–2205.
(45) Serianni, A. S.; Clark, E. L.; Barker, R. Carbon-13-Enriched Carbohydrates. Preparation of Erythrose, Threose, Glyceraldehyde, and Glycolaldehyde with 13C-Enrichment in Various Carbon Atoms. Carbohydr. Res. 1979, 72 (C), 79–91.
(46) Butlerow, A. Bildung Einer Zuckerartigen Substanz Durch Synthese. Justus Liebigs Ann. Chem. 1861, 120 (3), 295–298.
(47) Foster, G. W. A. XCV.—The Action of Light on Potassium Ferrocyanide. J. Chem. Soc. Trans. 1906, 89 (912), 912–920.
(48) Codere, F.; Dixon, D. G. Modeling the Cyanide Heap Leaching of Cupriferous Gold Ores. Hydrometallurgy 1999, 52 (2), 151–175.
(49) Sauer, M. C.; Crowell, R. A.; Shkrob, I. A. Electron Photodetachment from Aqueous Anions. I. Quantum Yields for Generation of Hydrated Electron by 193 and 248 Nm Laser Photoexcitation of Miscellaneous Inorganic Anions. J. Phys. Chem. A 2004, 108 (25), 5490–5502.
(50) Streecker, A. Ueber Einen Neuen Aus Aldehyd - Ammoniak Und Blausäure Entstehenden Körper. Ann. der Chemie und Pharm. 1854, 91 (3), 349–351.
(51) Schlesinger, G.; Miller, S. L. Equilibrium and Kinetics of Glycolaldehyde Formation in Aquous Solution. J. Am. Chem. Soc. 1973, 95 (11), 3729–3735.
(52) Shapiro, R. Prebiotic Ribose Synthesis: A Critical Analysis. Origins Life Evol. Biospheres 1988, 18 (1–2), 71–85.
(53) Ranjan, S.; Todd, Z. R.; Sutherland, J. D.; Sasselow, D. D. Sulfide Anion Concentrations on Early Earth for Surfacial Origins-of-Life Chemistry. Astrobiology 2018, 18 (8), 1023–1040.
(54) Yaroshovsky, A. A. Abundances of Chemical Elements in the Earth’s Crust. Geochim. Int. 2006, 44 (1), 48–55.
(55) Marion, G. M.; Kargel, J. S.; Crowley, J. K.; Catling, D. C. Sulphite-Sulphide-Sulfate-Carbonate Equilibria with Applications to Mars. Icarus 2013, 225 (1), 342–351.
(56) Chawla, O. P.; Arthur, N. L.; Fessenden, R. W. An Electron Spin Resonance Study of the Photolysis of Aqueous Sulfitic Solutions. J. Phys. Chem. 1973, 77 (6), 772–776.
(57) Keefe, A. D.; Miller, S. L. Was Ferrocyanide a Prebiotic Reagent? Origins Life Evol. Biospheres 1996, 26 (2), 111–129.
(58) Shrom, M.; Stein, G. Excited State Chemistry of the Ferrocyanide Ion in Aqueous Solution. I. Formation of the Hydrated Electron. J. Chem. Phys. 1971, 55 (7), 3372–3378.
(59) Lancaster, J. M.; Murray, R. S. The Ferricyanide-Sulphite Reaction. J. Chem. Soc. A 1971, 0, 2755–2758.
(60) Murray, R. S. An Intermediate in the Reaction between Ferrocyanide and Sulphite in Aqueous Solution. Chem. Commun. 1968, 14, 824.
(61) Wang, Z., Strecker Synthesis. Comprehensive Organic Name Reactions and Reagents; John Wiley & Sons, Inc.; Hoboken, NJ, 2010; pp 2710–2717.
(62) Shapiro, R.; DeFate, V.; Welcher, M. Deamination Cytosine in Nucleotides Revisited. Ribonucleotides: Photoanomerisation of Cytosine Nucleosides and Their Phosphate Derivatives Involving Photoanomerization. Nat. Chem. 2012, 4 (1), 1028–1032.
(63) Pascoe, D. J.; Ling, K. B.; Cockroft, S. L. The Origin of Chalcogen-Bonding Interactions. J. Am. Chem. Soc. 2017, 139 (42), 15160–15167.
(64) Rios, A. C.; Tor, Y. On the Origin of the Canonical Nucleobases: An Assessment of Selection Pressures across Chemical and Early Biological Evolution. Isr. J. Chem. 2013, 53 (6–7), 469–483.
(65) Sanchez, R. A.; Orgel, L. E. Studies in Prebiotic Synthesis. V. Synthesis and Photoanomerization of Pyrimidine Nucleosides. J. Mol. Biol. 1970, 47 (3), 531–543.
(66) Powner, M. W.; Anastasi, C.; Crowe, M. A.; Powner, M. W.; Sutherland, J. D. Direct Assembly of Nucleoside Precursors from Two- and Three- Carbon Units. Angew. Chem., Int. Ed. 2006, 45 (37), 6176–6179.
(67) Shrom, M.; Stein, G. Excited State Chemistry of the Ferrocyanide Ion in Aqueous Solution. I. Formation of the Hydrated Electron. J. Chem. Phys. 1971, 55 (7), 3372–3378.
(68) Hein, J. E.; Tse, E.; Blackmond, D. G. A Route to Enantiopure RNA Precursors from Nearly Racemic Starting Materials. Nat. Chem. 2011, 3 (9), 704–706.
(69) Xu, J.; Tsanakopoulou, M.; Magnani, C. J.; Szabla, R.; Šponer, J. E.; Šponer, J.; Góra, R. W.; Sutherland, J. D. A Prebiotically Plausible Synthesis of Pyrimidine λ2-Ribonucleosides and Their Phosphate Derivatives Involving Photoanomerization. Nat. Chem. 2017, 9 (4), 303–309.
(70) Xu, J.; Tsanakopoulou, M.; Magnani, C. J.; Szabla, R.; Šponer, J. E.; Šponer, J.; Góra, R. W.; Sutherland, J. D. A Prebiotically Plausible Synthesis of Pyrimidine λ2-Ribonucleosides and Their Phosphate Derivatives Involving Photoanomerization. Nat. Chem. 2017, 9 (4), 303–309.
(71) Grosjean, H.; de Crécy-Lagard, V.; Marck, C. Deciphering Synonymous Codons in the Three Domains of Life: Co-Evolution with Specific TRNA Modification Enzymes. FEBS Lett. 2010, 584 (2), 252–264.
(72) Wang, Z., Norrish Type II Reaction. In Comprehensive Organic Name Reactions and Reagents; John Wiley & Sons, Inc.; Hoboken, NJ, 2010; pp 2067–2071.
(73) Heuberger, B. D.; Pal, A.; Del Frate, F.; Topkar, V. V.; Szostak, J. W. Replacing Uridine with 2-Thiouridine Enhances the Rate and Fidelity of Nonenzymatic RNA Primer Extension. J. Am. Chem. Soc. 2015, 137 (7), 2769–2775.
(74) Verlander, M. S.; Lohrmann, R.; Orgel, L. E. Catalysts for the Self-Polymerization of Adenosine Cyclic 2’3’-Phosphate. J. Mol. Evol. 1973, 2 (4), 303–316.
(75) Schoffstall, A. M. Prebiotic Phosphorylation of Nucleosides in Formamide. Origins Life 1976, 7 (4), 399–412.
(76) Lohrmann, R.; Orgel, L. E. Urea-Inorganic Phosphate Mixtures as Prebiotic Phosphorylating Agents. Science 1971, 171 (3970), 490–494.
(77) Xu, J.; Green, N. J.; Gibbard, C.; Krishnamurthy, R.; Sutherland, J. D. Prebiotic Phosphorylation of 2-Thiouridine Provides Either Nucleotides or DNA Building Blocks via Photoreduction. Nat. Chem. 2019, 11 (5), 457–462.
(78) Reichard, P. From RNA to DNA, Why so Many Ribonucleotide Reductases? Science 1993, 260 (5115), 1773–1777.
(79) Burton, A. S.; Lehman, N. DNA before Proteins? Recent Discoveries in Nucleic Acid Catalysis Strengthen the Case. Astrobiology 2009, 9 (1), 125–130.
(80) Freeland, S. J. Do Proteins Predate DNA? Science 1999, 286 (5440), 690–692.
(81) Xu, J.; Chmela, V.; Green, N. J. J.; Russell, D. A. A.; Janicki, M. J.; Góra, R. W. W.; Szabla, R.; Bond, A. D. D.; Sutherland, J. D. Selective Prebiotic Formation of RNA Pyrimidine and DNA Purine Nucleosides. Nature 2020, 582 (7810), 60–66.
(82) Roberts, S. J.; Szabla, R.; Todd, Z. R.; Stairs, S.; Bučar, D.-K.; Šponer, J.; Sasselov, D. D.; Powner, M. W. Selective Prebiotic Conversion of Pyrimidine and Purine Anhydronucleosides into Watson-Crick Base-Pairing Arabinon-Furanosyl Nucleosides in Water. Nat. Commun. 2018, 9 (1), 4073.
(83) Mancini, R. L.; McKay, C. P. The Evolution of Nitrogen Cycling. Origins Life Evol. Biospheres 1988, 18 (4), 311–325.
(84) Wang, Z.; Passerini Reaction. In Comprehensive Organic Name Reactions and Reagents; John Wiley & Sons, Inc.; Hoboken, NJ, 2010; pp 2121–2125.
(85) Sutherland, J. D.; Mullen, L. B.; Buchet, F. F. Potentially Prebiotic Passerini-Type Reactions of Phosphates. Synlett 2008, 2008 (14), 2161–2163.
(86) Mullen, L. B.; Sutherland, J. D. Simultaneous Nucleotide Activation and Synthesis of Amino Acid Amides by a Potentially Prebiotic Multi-Component Reaction. Angew. Chem., Int. Ed. 2007, 46 (42), 8063–8066.
(87) Rocha, R. O.; Rodrigues, M. O.; Neto, B. A. D. Review on the Ugi Multicomponent Reaction Mechanism and the Use of Fluorescent Derivatives as Functional Chromophores. ACS Omega 2020, 5 (2), 972–979.
(88) Szostak, J. W. The Eightfold Path to Non-Enzymatic RNA Replication. J. Syst. Biol. 2012, 3 (1), 2.
(89) Bowler, F. R.; Chen, C. K. W.; Duffy, C. D.; Gerland, B.; Islam, S.; Powner, M. W.; Sutherland, J. D.; Xu, J. Prebiotically Plausible Oligoribonucleotide Ligation Facilitated by Chemoselective Acetylation. Nat. Chem. 2013, 5 (5), 383–389.
(90) Liu, Z.; Wu, L. F.; Xu, J.; Bonfoh, C.; Russell, D. A.; Sutherland, J. D. Harnessing Chemical Energy for the Activation and Joining of Prebiotic Building Blocks. Nat. Chem. 2020, 12 (11), 1023–1028.
(91) Bonfoh, C.; Russell, D. A.; Green, N. J.; Mariani, A.; Sutherland, J. D. Activation Chemistry Drives the Emergence of Functionalised Protocells. Chem. Sci. 2020, 11 (39), 10688–10697.
(92) Vargese, C.; Carter, J.; Yegge, J.; Krivjansky, S.; Settle, A.; Kropp, E.; Peterson, K.; Pienek, W. Efficient Activation of Nucleoside Phosphoramidites with 4,5-Dicyanomimidazole during Oligonucleotide Synthesis. Nucleic Acids Res. 1998, 26 (4), 1046–1050.
(97) Sagan, C. Ultraviolet Selection Pressure on the Earliest Organisms. J. Theor. Biol. 1973, 39 (1), 195–200.

(98) Cleaves, H. J.; Miller, S. L. Oceanic Protection of Prebiotic Organic Compounds from UV Radiation. Proc. Natl. Acad. Sci. U. S. A. 1998, 95 (13), 7260–7263.

(99) Bucher, D. B.; Kufner, C. L.; Schlueter, A.; Carell, T.; Zinth, W. UV-Induced Charge Transfer States in DNA Promote Sequence Selective Self-Repair. J. Am. Chem. Soc. 2016, 138 (1), 186–190.

(100) Todd, Z. R.; Fahrenbach, A. C.; Magnani, C. J.; Ranjan, S.; Björkbom, A.; Szostak, J. W.; Sasselov, D. D. Solved-Electron Production Using Cyanocuprates Is Compatible with the UV-Environment on a Hadean-Archaean Earth. Chem. Commun. 2018, 54 (9), 1121–1124.

(101) Ranjan, S.; Sasselov, D. D. Influence of the UV Environment on the Synthesis of Prebiotic Molecules. Astrobiology 2016, 16 (1), 68–88.

(102) Todd, Z. R.; Szabla, R.; Szostak, J. W.; Sasselov, D. D. UV Photostability of Three 2-Aminoazoles with Key Roles in Prebiotic Chemistry on the Early Earth. Chem. Commun. 2019, 55 (70), 10388–10391.

(103) Janicki, M. J.; Roberts, S. J.; Šponer, J.; Powner, M. W.; Góra, R. W.; Szabla, R. Photostability of Oxazoline RNA-Precursors in UV-Rich Prebiotic Environments. Chem. Commun. 2018, 54 (95), 13407–13410.

(104) Todd, Z. R.; Szostak, J. W.; Sasselov, D. D. Shielding from UV Photodamage: Implications for Surficial Origins of Life Chemistry on the Early Earth. ACS Earth Sp. Chem. 2021, 5 (2), 239–246.

(105) Eschenmoser, A.; Loewenthal, E. Chemistry of Potentially Prebiological Natural Products. Chem. Soc. Rev. 1992, 21 (1), 1.

(106) Zahnele, K. J.; Lupu, R.; Catling, D. C.; Wogan, N. Creation and Evolution of Impact-Generated Reduced Atmospheres of Early Earth. Planet. Sci. J. 2020, 1 (1), 11.

(107) Toner, J. D.; Catling, D. C. Alkaline Lake Settings for Concentrated Prebiotic Cyanide and the Origin of Life. Geochim. Cosmochim. Acta 2019, 260, 124–132.

(108) Liu, Z.; Wu, L. F.; Bond, A. D.; Sutherland, J. D. Photoredox Chemistry in the Synthesis of 2-Aminoazoles Implicated in Prebiotic Nucleic Acid Synthesis. Chem. Commun. 2020, 56 (88), 13563–13566.

(109) Schmitt-Kopplin, P.; Gabelica, Z.; Gougeon, R. D.; Fekete, A.; Kanawati, B.; Harir, M.; Gebefuegi, I.; Eckel, G.; Hertkorn, N. High Molecular Diversity of Extraterrestrial Organic Matter in Murchison Meteorite Revealed 40 Years after Its Fall. Proc. Natl. Acad. Sci. U. S. A. 2010, 107 (7), 2763–2768.

(110) Szostak, J. W. Systems Chemistry on Early Earth. Nature 2009, 459 (7244), 171–172.

(111) Miller, S. L.; Bada, J. L. Submarine Hot Springs and the Origin of Life. Nature 1988, 334 (6183), 609–611.