

PREBIOTIC CHEMISTRY

Mist and replication

The transition from chemistry to evolvable molecular systems is at the core of origins of life studies. Now, the acidic dew–liquid water dynamic cycling inside simulated Hadean rock pores is found to possibly provide a confined environment for strand separation, replication, mutation, and the evolution of nucleic acids.

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The transition from chemistry to biology is thought to have happened relatively soon after the Earth–Moon system was formed. This period, the Hadean eon, likely had many energy sources available to facilitate the prebiotic reactions that eventually gave rise to life on Earth.

One especially pertinent prebiotic event central to life's origins involves the formation of informational biopolymers from monomeric precursors. Many studies have shown the formation of key precursor molecules and canonical nucleobases from prebiotic feedstock molecules under Hadean conditions^{1,2}. These are fundamental for the subsequent formation of informational and functional molecules capable of supporting Darwinian evolution, such as RNA or proto-RNA. Writing in *Nature Physics*, Ianeselli and colleagues³ report intriguing experimental results that may represent an important step towards understanding how these molecules might subsequently have evolved.

A fundamental challenge in this area is centred around the many competing requirements that must be satisfied in order to promote the different chemical processes that ultimately favour the replication of these polymers. Once established, these molecules would have required not only efficient catalysis for replication, but also a process for the cyclic separation and re-hybridization of complementary strands. Making this a challenge, the physico-chemical conditions that favour replication reactions typically disfavour the melting of oligonucleotides.

Strand separation might be promoted by changes in pH, wet–drying cycles, salt concentration gradients, or temperature oscillations^{4,5}. Yet, these events can be destabilizing or all together deleterious for the replicating molecule. For example, high pH degrades RNA, low salts can be adverse to catalysis, and loss of informational moieties can happen readily under simulated prebiotic conditions⁶.

A plausible theory for the prebiotic appearance and evolution of Darwinian

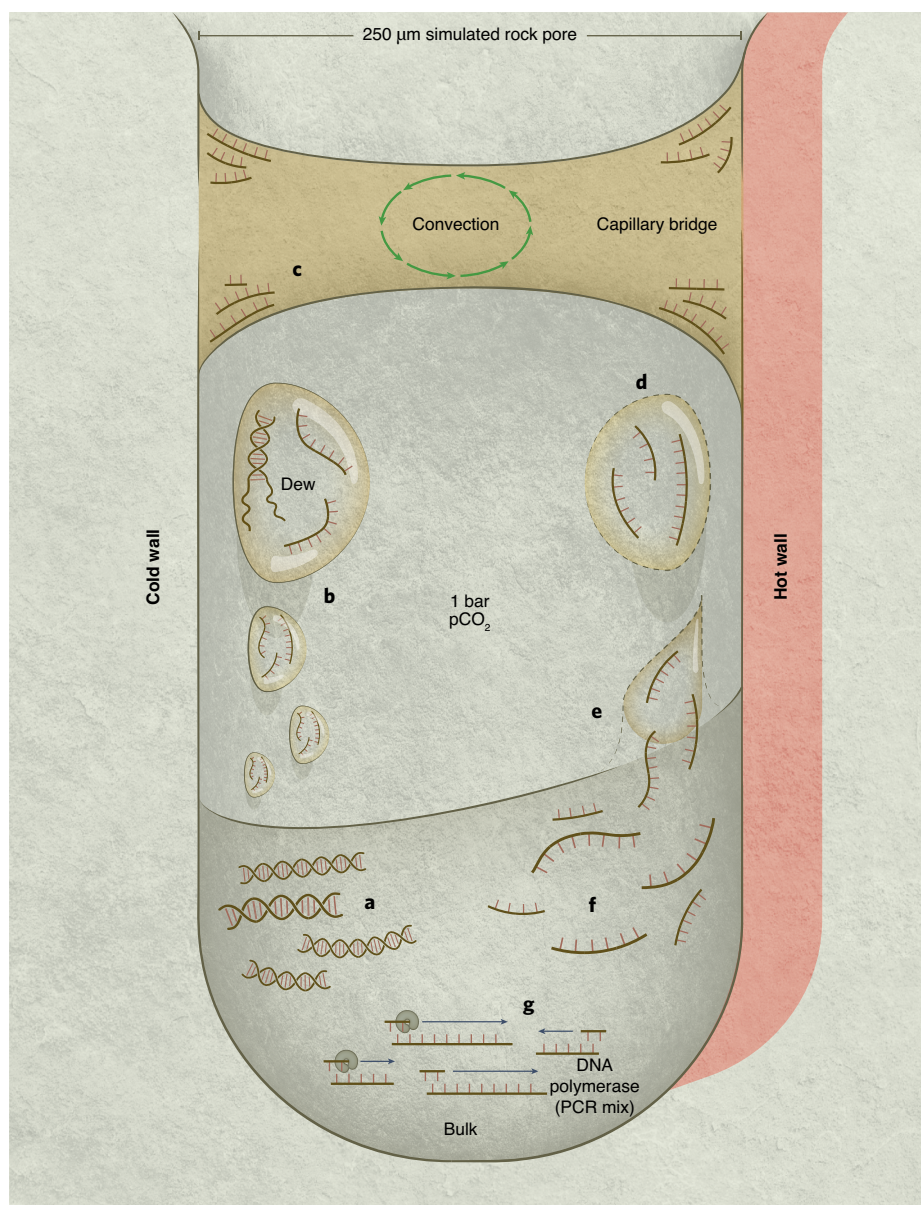


Fig. 1 | Differentially heated simulated rock pores create a prebiotic thermocycler. a–d, Double-stranded nucleic acid molecules in the bulk at near neutral pH (a) are trapped inside low-salt, low-pH dew droplets (b,d) and denatured into single strands. A capillary bridge connects the hot and cold walls of the pore, while thermal convection allows for continuous dew cycling (c). **e–g**, Coalescence of the dew into the bulk (e) allows for release of single-stranded nucleic acids (f) into the bulk, where re-amplification creates new double-stranded molecules (g). PCR, polymerase chain reaction. Image courtesy of Souradeep Das.

systems must therefore provide a setting that minimizes the spontaneous cleavage of polymers at high temperatures, while still facilitating conditions required for catalysis. Importantly, this setting should also allow for compartmentalization so as to avoid the diffusion of the template strands and the oligomerizing monomers.

Ianeselli and colleagues have now begun to show that strand separation, replication, mutation and evolution of nucleic acid sequences can be achieved in the dew–liquid water dynamic cycling inside simulated rock pores at Hadean CO₂ pressure.

Their study builds on previous work in which they developed a lab model of a Hadean rock pore subjected to temperature gradients. The pore is composed of a chamber placed between two plates, which are differentially heated to provide a gradient. This can be partially filled with solutions containing various types of molecules, ultimately resembling a volcanic rock pore subjected to flooding in a hydrothermal setting. When a thermal gradient is applied across this simulated pore, a ‘dew chamber’ is created where water cycles from liquid to gas phases between the two differentially heated walls, providing the dissolved molecules different physico-chemical environments in the same enclosed space.

Using this ingenious contraption, Ianeselli and co-workers first observed accumulation of nucleotides and other prebiotic molecules at the water–gas interface, preferential amplification of longer DNA oligomers in thermal gradients, and strand separation in thermally induced salt oscillations^{4,7,8}.

The group’s latest work takes this a step further, by introducing in the chamber acidic CO₂ pressure conditions resembling those likely present on Hadean Earth⁹. Atmospheric CO₂ was introduced into the chamber at pressures between 0.4 mbar (ambient pCO₂) and 1 bar. Beside the previously reported accumulation of oligomers at the gas–water interface and their sequestration inside the dew, the authors detected a substantial drop in pH between the liquid phase and the droplets (from ~pH 5.8 to ~pH 4.0) at 1 bar CO₂. Along with the depletion of salts from the droplets, the drop in pH triggers the denaturation of double-stranded DNA

and RNA molecules as measured using fluorescence resonance energy transfer (FRET). In contrast, high salt concentrations and near neutral pH in the bulk phase stabilize the double-stranded conformation.

The authors then test these different conditions for their ability to promote denaturing and renaturing cycles typical of amplification reactions. Double-stranded DNA was thus introduced into the chamber along with *Taq* DNA polymerase, dNTPs and buffers, and a heat difference of 15 °C was applied between the two plates. Remarkably, the authors report efficient amplification of DNA when 1 bar CO₂ pressure is applied, but no amplification when pCO₂ is ambient, as there is no pH gradient between the bulk and the dew.

This amplified material was then subjected to nanopore sequencing¹⁰, and the authors found that the replicated DNA consistently grows in size and mutates in sequence, converging towards highly AT-rich sequences. This preference can be explained by the specific conditions of the chamber, which allow for those sequences that can denature easily to amplify exponentially, at the expense of those forming stronger duplexes (GC-rich sequences). This was confirmed by the authors with a simulation study of the FRET landscape for an evolving DNA sequence.

Hence, Ianeselli et al. report a differentially heated rock pore that is capable of generating salt and pH gradients by virtue of convection flows and adsorption of CO₂ from the Hadean atmosphere. This system allows for efficient RNA melting 30 K below the bulk melting temperature, and efficient (enzymatic) amplification and mutation of long DNA molecules with cycled strand separation, essentially acting as a bona fide ‘prebiotic thermocycler’ (Fig. 1).

Of course, still missing from the picture is the appearance in the simulated rock pore of a truly prebiotic, self-sufficient macromolecular system capable of undergoing Darwinian evolution.

Similarly, with Teflon making up the walls of the CO₂ dew chamber, this work does not take into account (yet) the potential catalytic power that might be embedded in the surface of prebiotic volcanic rock pores, and, in general, the contribution of geochemically active surfaces¹¹.

The presence of such rock pores at different locations, such as in proximity to hydrothermal vent systems or in confined aquifers above the crust, would likely influence the choice of biomolecules that get selected for participating in condensation reactions like polymerization. We cannot exclude the possibility that polymers with very different compositions (for example nucleic acids, peptides or xenobiologics¹²) might have formed and perhaps replicated concomitantly in similar rock pores, but within different geochemical settings. Moreover, dense liquid droplets of macromolecules known as coacervates might also have come into play and contributed to the amplification of this prebiotic system, as recently reported¹³ by the same group in *Nature Chemistry*.

Ianeselli and co-workers will undoubtedly continue to build on these exciting results, aiming to provide a detailed and complete picture of what the emergence of replicating prebiotic systems in Hadean rock pores could have actually looked like. □

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Competing interests

The authors declare no competing interests.