



Hybrid organic–inorganic structures trigger the formation of primitive cell-like compartments

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Alkaline hydrothermal vents have become a candidate setting for the origins of life on Earth and beyond. This is due to several key features including the presence of gradients of temperature, redox potential, pH, the availability of inorganic minerals, and the existence of a network of inorganic pore spaces that could have served as primitive compartments. Chemical gardens have long been used as experimental proxies for hydrothermal vents. This paper investigates a set of prebiotic interactions between such inorganic structures and fatty alcohols. The integration of a medium-chain fatty alcohol, decanol, within these inorganic minerals, produced a range of emergent 3 dimensions structures at both macroscopic and microscopic scales. Fatty alcohols can be considered plausible prebiotic amphiphiles that might have assisted the formation of protocellular structures such as vesicles. The experiments presented herein show that neither chemical gardens nor decanol alone promote vesicle formation, but chemical gardens grown in the presence of decanol, which is then integrated into inorganic mineral structures, support vesicle formation. These observations suggest that the interaction of fatty alcohols and inorganic mineral structures could have played an important role in the emergence of protocells, yielding support for the evolution of living cells.

origin of life | vesicles | inorganic mineral surfaces | surface chemistry | alkaline hydrothermal vents

One of the leading models for the emergence of life on earth is the alkaline hydrothermal vent theory (1–5). Off-axis hydrothermal vents (comparable to the well-known Lost City vent field) are focusing centers for various thermodynamic gradients that can drive a wide range of chemical processes (6–9). Cold, CO₂-rich, oxidized sea water as present in the Hadean (10–12), percolates into the porous, mineral environs of the vents. Within the vent chimneys lie networks of microscopic pores, through which the fluid is permeating (3). The seawater eventually circles back to rejoin the ocean, passing through the growing vent chimneys and emerging very different in composition and temperature (13–15). Accordingly to the alkaline vent theory, an array of prebiotic reactions, exploiting disequilibria and the presence of small-molecule precursors took place, producing biomolecules such as small peptides (16–20). In the meantime, the rock-water reaction known as serpentinization could have released H₂ in the midst of such vent structures, enabling reducing reactions such as alkane polymerization (3) and ketone reduction to alcohols (21). We can surmise that the synthesis of many prebiotic molecules would have occurred in the presence of hydrothermal vents, that created favorable conditions for the emergence of primitive biochemistry (9, 22, 23).

Alongside molecular synthesis, a key question is the spatial organization (if any) of these complex distributions of compounds. In fact, vent scenarios provide natural mechanisms by which prebiotic molecules could have self-organized toward the first compartmentalized protocells (24, 25), and recent work by Jordan et al. (5, 26) demonstrated the stable self-assembly of protocellular structures under hydrothermal conditions. In contemporary research on protocells, fatty acids are a primary focus (27, 28), and it was demonstrated that RNA can be encapsulated (29, 30) and copied (31) inside fatty acid vesicles. Furthermore, in the context of origins of life scenarios, the stabilizers of early phosphate amphiphile membranes were likely mixtures of mid- to long-chain fatty acids and alcohols (32–34), and medium- to long-chain alcohols have been shown to increase the stability of primitive membranes (35–39). Hydrothermal vents are also likely to have been a source of fatty acids and medium-chain alcohols derived from alkanes (40, 41).

This growing body of results is suggestive of a key role for fatty alcohols and their interactions with fatty acids in hydrothermal vent origin scenarios. In this work, we

Significance

Hydrothermal vents have long been suggested as an ideal location where abiogenesis could have occurred. Simultaneously, a large volume of research has explored how the first cell membranes may have arisen, leading to the evolution of the first living cells. The present study demonstrates the integration of these two parallel bodies of work in the origins of life. The inclusion of organic molecules in vent analog systems led to the creation of new and unique 3D surfaces, which support the self-assembly of vesicles. This demonstrates that the presence of organic chemistry can modify the inorganic surface architectures, which together lead to the promotion of primitive cell-like compartments.

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will focus specifically on one fatty alcohol and three fatty acids in order to keep the system sufficiently simple with a manageable set of control parameters. In particular, we endeavored to explore whether a medium-chain fatty alcohol could be integrated into inorganic mineral vent systems. Chemical gardens have a widely cited role as an effective proxy for laboratory studies in this area; thus, we employed this system in the present work. Commonly used prototypes of Lost City vent field include aragonite, brucite, calcite, and silicate (42–44). Given the presence of calcium carbonate and silicate minerals in Lost City Vents, we chose to reproduce calcium silicate gardens.

As an amphiphilic, 10-carbon chain alcohol, decanol is known to be a weak surfactant with low solubility in water (45). It is the shortest chain length alcohol capable of phase separation in the presence of water and has sufficient chain length to incorporate into fatty acid bilayer membranes and modify their properties without destroying them (35). These properties make decanol an interesting candidate to study as a potential component in prebiotic membranes. Given the plausible presence and importance of these alcohol molecules in ancient alkaline vent scenarios, we analyzed the feasibility of interactions between decanol, alkaline hydrothermal vents, and fatty acids using chemical gardens as a proxy. A range of emergent structures, at both microscopic and macroscopic scales, were observed, and fatty acid vesicle formation was triggered only in the presence of the crystal garden grown in the presence of decanol.

1. Results

A. Macroscopic and Microscopic Effects of Decanol on Chemical Gardens: Vertical and Horizontal Systems. Macroscopic alterations of two main variants of the chemical garden were grown and observed in vertical and horizontal systems. The systems studied are CG in the case of a typical chemical garden and CGD in the case of the chemical garden grown in the presence of decanol. Decanol was added in CGD systems previously to CaCl_2 addition.

Calcium chloride seeds when added to a sodium silicate solution fall to the bottom of the glass vial where the silicate solution is previously loaded and create chemical gardens that grow from the seeds upward (Fig. 1*A*, CG). Fig. 1*B* and *C* (CGD) show two alternative systems where 1-decanol is added to the sodium silicate solution. In Fig. 1*B*, decanol is gently layered on top of the silicate solution, and CaCl_2 seeds are dropped upon it. The seeds tend to stay at the decanol–silicate interface where they form a solid barrier. In Fig. 1*C*, decanol and silicate are vigorously shaken before CaCl_2 seeds are dropped upon the created emulsion. Some seeds became trapped at the interface, some fell to the bottom of the tube or stopped part way through the decanol silicate emulsion, depending on the seed's weight and on the density gradient. In both systems with decanol, the seeds form distinct macroscopic structures, varying between one another and in comparison with the CG system. The CGD structures resemble ceiling-like barriers and web-like structures (*SI Appendix, Video S1*).

Further analysis of the structures was performed in a horizontal observation system. Sodium silicate solution was placed in each well of a 6-well plate. Decanol was added to three of the wells, and calcium chloride powder was added manually to all 6 wells. Examples of the obtained systems are represented in Fig. 1*D* (CG) and *E* (CGD). The evolving structures in these two different systems (CG & CGD) look markedly different

(*SI Appendix, Video S2*). The crystals localized at the interface between silicate solution and decanol in CGD systems showed random motion. We report in *SI Appendix* a pixel intensity plot that portrays the typical observational differences between CG and CGD horizontal systems (*SI Appendix, Fig. S1*). We show that CGD systems are macroscopically different from CG systems using optical images, video analysis, and pixel analysis. In the CG system (bottom row, Video 2), the calcium chloride powder fell to the bottom of the well and began to generate a chemical garden, starting from where it was dropped and with the auxiliary help of the well's wall interface. In CGD systems (top row), some calcium chloride powder was trapped at the interface, and some fell to the bottom of the well. The powder trapped at the interface showed some random motion, and the powder that reached the bottom of the well slowly reacted. We then shifted to the microscopic scale and analyzed CG and CGD systems after 1 d and after 1 mo of incubation using scanning electron microscopy (SEM). After 1 d and after 1 mo, the decanol phase was still clearly phase separated from the silicate water phase and transparent. There are clear differences between structures formed without (Fig. 1*F*) and with decanol (Fig. 1*G*) over 1 d. Samples formed without decanol are similar to those typically observed in chemical gardens, mainly composed of a sodium silicate matrix with garden-like features as well as spike-like structures. Samples incubated with decanol instead show more orthogonal structures with some crystals and rosette formations. Systems incubated for 1 mo (Fig. 1*H* and *I*) show some similar variations and structures to those incubated for 1 d (Fig. 1*F* and *G*). As above, samples left to react without decanol evolved similarly to typical chemical gardens, and metal ion jets likely created the observable small bubble-like structures (Fig. 1*H*). In contrast, the systems incubated with decanol produced squared protrusions and rectangular structures, demonstrating the long-term influence of decanol addition on the microscopic structure of chemical gardens (Fig. 1*J*). Thus, we deduce that on the microscopic scale, decanol strongly influences the morphology of the mineral inorganic surfaces created from the incubation of sodium silicate with calcium chloride.

B. Chemical Composition Effects of Decanol on Chemical Gardens: X-ray Photoelectron Spectroscopy and Scanning Electron Microscopy Coupled with Energy Dispersive X-ray Spectroscopy. After scanning these diverse structures using SEM, X-ray photoelectron spectroscopy (XPS) was used to further analyze the inorganic mineral surface composition. Three samples of type CG and three samples of type CGD were analyzed. Samples were left to react for 1 d before analysis.

We first analyzed the overall crystal surface composition using long-range XPS spectra. The long-range XPS spectra in Fig. 2*A* show the presence of sodium (Na1s), silicon (Si2p), oxygen (O1s), chlorine (Cl2p), carbon (C1s), and calcium (Ca2p). This analysis shows expected compositions for all the samples. All of them are characterized by one Na1s thin peak from Na–O bonds, and one broad peak in the Si2p profile at 108.45 eV attributed to Si–O bonds (102 eV) and Si–O–Si bonds (103 eV), typical of sodium silicates (46). The O1s profile is characterized by an asymmetric band and two types of oxygen in the silicate matrix: bridging O atoms (BO) connecting two Si oxide tetrahedra covalently and nonbonding O atoms (NBO) bonded to Si but also ionically connected to another cation (in this case, Na or Ca). Furthermore, lineshape fitting was optimized accounting for the contribution of a third component in the deconvolution analysis, with an energy region typical of M–OH bonds (lineshape