

Hypothesis

Ancient Yet Alive: Stable Molecular Networks and Precellular Polymer Consortia as Frameworks for Alternative Hereditary Systems Before LUCA

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Abstract

The origin of life is commonly framed within the RNA World hypothesis, yet increasing evidence suggests that early evolution may have involved a broader diversity of hereditary systems than those preserved in modern biology. Alternative informational polymers, compositional inheritance, autocatalytic networks, and compartmentalized molecular communities have all been proposed as mechanisms that may have preceded modern genome-based heredity. Building upon these concepts, we propose that the fundamental unit of early evolution may have been neither the gene nor the organism, but persistent molecular networks capable of maintaining organizational continuity through time. We define Stable Molecular Networks (SMNs) as interacting systems of informational polymers, catalytic oligomers, metabolites, compartments, and environmental feedback processes that maintain continuity despite continual molecular turnover. We further propose that compartmentalized SMNs could form molecular ecosystems termed Precellular Polymer Consortia (PPCs), in which heredity emerges from network organization rather than genome replication alone. Within PPCs, selective interactions, molecular memory, ecological feedback, and distributed information exchange may generate lineage-like evolutionary processes that we term precellular speciation. To connect these concepts to extant biology, we examine archaeal and bacterial systems that preserve organizational principles potentially relevant to early evolution, including CRISPR-Cas adaptive memory systems, ancient RNA-based molecular machines such as the ribosome and RNase P, hydrothermal vent archaea, chemolithotrophic microorganisms, syntrophic consortia, and complex microbial communities. Although these systems are not direct descendants of precellular networks, they provide experimentally accessible examples of molecular memory, distributed information processing, protometabolism, ecological cooperation, and system-level organization. Finally, we propose PLURIBUS (Planetary Liquid Universal Polymer Identification By Ultrasensitive Sequencing) as a framework for discovering noncanonical informational polymers, novel RNA modifications, and alternative hereditary systems within Earth's microbial biosphere, particularly among poorly characterized archaeal and extremophile communities. Knowledge gained from these environments may ultimately support the development of more general life-detection strategies for planetary exploration. Together, these perspectives suggest that extant microbial ecosystems provide valuable experimental windows into evolutionary processes that may have preceded modern cells, genomes, and the Last Universal Common Ancestor (LUCA).



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1. Introduction

Understanding how life emerged from nonliving matter remains one of the most fundamental challenges in science. Over the past several decades, the RNA World hypothesis has become the dominant framework for explaining this transition [1–6]. The discovery of catalytic RNAs by Altman [7] and Cech [8–10], advances in prebiotic nucleotide synthesis [11,12], and laboratory evolution of ribozymes [13,14] have demonstrated that RNA can function both as a hereditary molecule and as a catalyst. Together, these discoveries provide strong evidence that RNA-based systems likely played important roles during early evolution.

Despite these successes, several fundamental questions remain unresolved. It is unclear whether canonical RNA was the first hereditary polymer [15,16], whether alternative hereditary systems existed prior to RNA-based biology [17,18], and how the transition occurred from simple chemical mixtures to evolving molecular communities capable of persistence, heredity, and diversification [17–19]. Modern biology relies almost exclusively on DNA, RNA, proteins, and lipid membranes, but these molecules may represent only a small surviving subset of the molecular diversity explored during early planetary evolution.

Several observations support a broader view of early heredity. Laboratory studies have demonstrated that alternative hereditary polymers—including peptide nucleic acids (PNA) [20], threose nucleic acids (TNA) [21], glycol nucleic acids (GNA) [22], and other xeno nucleic acids (XNAs) [23]—can store information and participate in template-directed interactions. Contemporary RNA biology itself exhibits extensive chemical diversity, with hundreds of naturally occurring nucleotide modifications cataloged in the MODOMICS database [24,25]. In addition, samples returned from the asteroids Bennu and Ryugu contain amino acids, nucleobases, sugars, ammonia-rich compounds, and complex organic matter [26–28], demonstrating that sophisticated organic chemistry occurs naturally beyond Earth.

While studies of prebiotic chemistry provide insight into possible pathways toward life, extant microbial systems may also illuminate early evolutionary transitions. CRISPR-Cas adaptive memory systems [29–31], ancient RNA-based molecular machines such as the ribosome and RNase P [32–34], thermophilic archaea [35], chemolithotrophic bacteria [36–38], and hydrothermal vent communities [39–41] provide living examples of molecular memory, information processing, protometabolism, and ecological organization. Although modern microorganisms are highly evolved rather than primitive relics, they offer experimentally accessible systems for investigating organizational principles that may have contributed to the emergence of heredity before the Last Universal Common Ancestor (LUCA). The fundamental unit of early evolution may therefore have been neither the gene nor the organism, but the stable molecular network itself.

A major challenge in origins-of-life research is therefore to define heredity more broadly than genome replication alone. Several theoretical frameworks have emphasized compositional inheritance, autocatalytic networks, dynamic kinetic stability, communal evolution, and distributed molecular organization as potential precursors to modern genome-based heredity [42–49]. Building upon these concepts, we propose that Stable Molecular Networks (SMNs)—persistent interacting systems of informational polymers, catalytic oligomers, compartments, metabolites, and environmental feedback—may represent an

intermediate organizational stage between planetary chemistry and cellular life. In this framework, continuity is maintained not by individual molecules alone but by persistent patterns of interaction among molecular components.

We further propose that compartmentalized molecular ecosystems, termed Precellular Polymer Consortia (PPCs), may have arisen from Stable Molecular Networks before the emergence of modern cells. Within PPCs, information storage, amplification, ecological interactions, and selective communication among molecular communities could generate lineage-like evolutionary processes prior to biological reproduction. To explore this possibility, we examine alternative hereditary systems, contemporary molecular memory pathways, extant archaeal and microbial systems, and their implications for astrobiology and future life-detection strategies.

These possibilities also motivate new strategies for detecting unknown hereditary chemistries in extant microbial ecosystems. Throughout this review, we propose that discoveries from extreme archaeal and microbial environments may provide the foundation for PLURIBUS (Planetary Liquid Universal Polymer Identification By Ultrasensitive Sequencing), a framework for identifying noncanonical informational polymers and molecular networks on Earth and beyond. Figure 1 summarizes the conceptual framework linking planetary chemistry, alternative hereditary systems, Stable Molecular Networks, Precellular Polymer Consortia, and the emergence of cellular life before LUCA. Collectively, these observations suggest that the fundamental unit of early evolution may have been neither the gene nor the organism, but persistent molecular networks capable of maintaining system persistence through time.

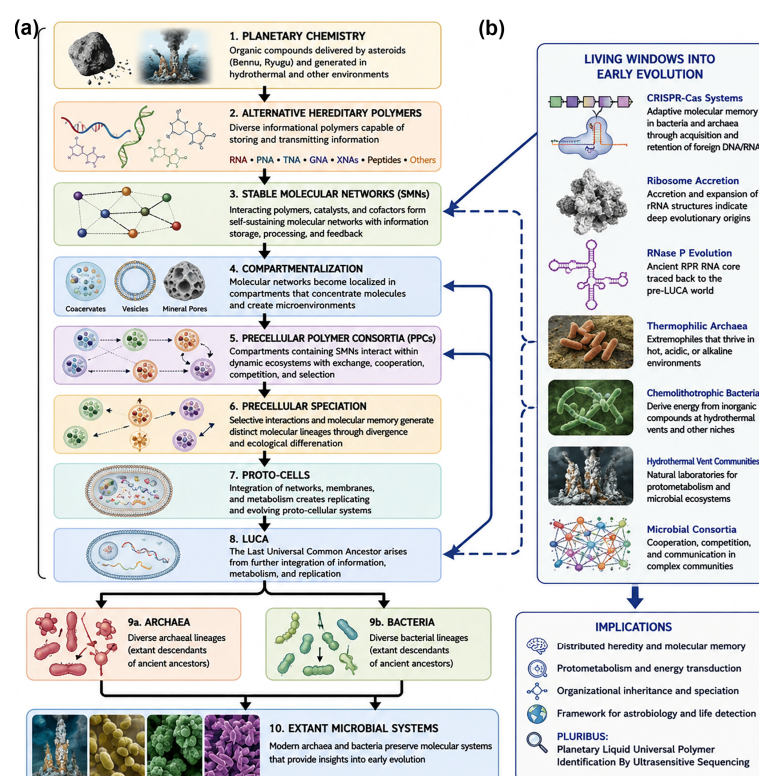


Figure 1. From planetary chemistry to extant microbial systems. (a) Conceptual model linking alternative hereditary polymers, Stable Molecular Networks (SMNs), Precellular Polymer Consortia (PPCs), precellular speciation, proto-cells, LUCA, and modern archaeal and bacterial lineages. (b) Extant microbial systems that may provide insights into early evolution, including CRISPR-Cas systems, ribosomes, RNase P, thermophilic archaea, chemolithotrophic bacteria, hydrothermal vent communities, and microbial consortia. These systems illustrate molecular memory, distributed heredity, protometabolism, and organizational continuity relevant to pre-LUCA evolution.

2. Materials and Methods

This article is a hypothesis-driven narrative review that integrates findings from origins-of-life research, molecular evolution, microbiology, astrobiology, systems biology, and RNA biology. Studies from the literature were identified through searches of PubMed, Google Scholar, Web of Science, and related databases using combinations of the terms origin of life, RNA World, alternative hereditary systems, CRISPR-Cas, ribosome evolution, RNase P, autocatalytic networks, compositional inheritance, hydrothermal vents, thermophilic archaea, chemolithotrophic bacteria, microbial consortia, LUCA, and astrobiology. The review synthesizes concepts from alternative hereditary polymers, compositional inheritance, autocatalytic network theory, dynamic kinetic stability, microbial molecular memory systems, and evolutionary models of early life. Emphasis is placed on extant archaeal and bacterial systems that may provide experimentally accessible insights into network behaviors relevant to pre-LUCA evolution. Based on this synthesis, we develop the concepts of Stable Molecular Networks (SMNs) and Precellular Polymer Consortia (PPCs) as hypothetical intermediates linking planetary chemistry, early hereditary systems, and cellular life. These concepts are intended as heuristic frameworks that generate experimentally testable predictions of molecular ecosystems (Box 1) rather than definitive models of early evolution.

Box 1. Molecular Ecosystem.

A molecular ecosystem is a network of interacting informational polymers, catalytic molecules, metabolites, compartments, and environmental feedback processes capable of maintaining system persistence through time. Unlike modern biological organisms, molecular ecosystems need not possess genomes, cells, or biological reproduction. Their persistence arises from interactions among system components rather than replication of individual molecules. Within the framework proposed here, molecular ecosystems may range from prebiotic chemical systems to compartmentalized assemblies such as Stable Molecular Networks (SMNs) and Precellular Polymer Consortia (PPCs). Modern microbial communities, including hydrothermal vent ecosystems, microbial mats, and multispecies consortia, provide contemporary examples of how distributed interactions among diverse components can generate emergent system-level properties. Although molecular ecosystems are not proposed to be direct ancestors of modern microorganisms, extant archaeal and bacterial systems may preserve organizational principles—including molecular memory, cooperative interactions, resource exchange, compartmentalization, and ecological selection—that are relevant to understanding evolutionary transitions preceding LUCA.

3. Alternative Hereditary Systems Beyond the RNA World

3.1. *Why Early Life Was Probably Not Modern RNA*

More than 300 naturally occurring RNA modifications cataloged in MODOMICS demonstrate that contemporary RNA already occupies a broader chemical space than the canonical four nucleotides [24,25]. Furthermore, most of these modifications were identified from abundant RNAs isolated from cultivable organisms using mass spectrometry-based approaches. Consequently, current catalogs may represent only a fraction of the chemical diversity present within Earth's microbial biosphere, particularly among uncultured archaea and extremophiles.

3.1.1. Alternative Hereditary Polymers

Experimental studies have shown that numerous noncanonical nucleic acid systems can store information and participate in template-directed molecular interactions. Alternative backbones, sugars, and nucleobases can support complementary recognition and heredity-like processes (Table 1).

Table 1. Representative Alternative Hereditary Polymers and Their Potential Relevance to Early Hereditary Systems. Representative informational polymers that demonstrate the chemical flexibility of hereditary systems. Heredity storage, molecular recognition, complementary pairing, and heredity-like behavior are not unique to canonical RNA and DNA. The existence of multiple experimentally validated nucleic acid analogs suggests that early evolutionary systems—and potentially extraterrestrial life—may employ molecular architectures fundamentally different from those used by contemporary terrestrial organisms.

Polymer System	Backbone/Sugar	Key Informational Properties	Experimental Support	Potential Astrobiological Relevance	References
PNA (Peptide Nucleic Acid)	Peptide-like backbone	Strong base pairing, high chemical stability, resistant to enzymatic degradation	Template-directed information transfer demonstrated experimentally	Candidate alternative hereditary system in non-terrestrial environments	[20]
TNA (Threose Nucleic Acid)	Threose sugar	Simpler four-carbon sugar, capable of Watson–Crick pairing with RNA	Laboratory evolution and information exchange with RNA demonstrated	Potential pre-RNA or extraterrestrial informational polymer	[21]
GNA (Glycol Nucleic Acid)	Glycol backbone	Structurally simple, stable duplex formation	Synthetic heredity-like properties demonstrated	Illustrates that heredity is not unique to ribose-based systems	[22]
HNA (Hexitol Nucleic Acid)	Six-carbon sugar	Stable helical structures, capable of information storage and transfer	Evolved HNA aptamers and hereditary functions reported	Demonstrates viability of alternative sugar chemistries	[50]
ANA/FANA	Arabinose-based sugars	Alternative stereochemistry, hybridization with RNA and DNA	Functional nucleic acid analogs produced experimentally	Highlights chemical flexibility of informational systems	[51–53]
Expanded Genetic Alphabets (AEGIS, XNAs)	Variable	Utilize noncanonical nucleobases to increase coding capacity	Multiple synthetic systems demonstrated	Suggests extraterrestrial hereditary systems may use larger molecular alphabets	[23]
Modified RNA Systems	Ribose with chemically modified bases	Expanded catalytic and structural capabilities	Supported by >300 modifications cataloged in MODOMICS	Potential remnants of earlier molecular diversity	[24,25]
Hypothetical Extraterrestrial Informational Polymers	Unknown	May utilize alternative sugars, backbones, or nucleobases	Not yet observed directly	Possible hereditary systems in environments beyond Earth	This paper

The diversity of experimentally validated hereditary polymers raises an important practical question: how many additional informational chemistries remain undiscovered in nature? Most known nucleic acid analogues have been investigated in laboratory systems, whereas many archaeal and microbial lineages remain difficult to culture and molecularly characterize. Future sequencing technologies capable of detecting unknown polymers and chemical modifications directly may therefore reveal a substantially broader hereditary landscape than currently recognized.

Although none of these alternative polymers have been identified in extant organisms as primary hereditary systems, their experimental viability demonstrates that information storage and molecular recognition are not unique to RNA and DNA. Rather than viewing RNA as uniquely capable of supporting life, these findings suggest that hereditary behavior may emerge from a broader class of molecular systems. Such diversity is particularly relevant to astrobiology because extraterrestrial hereditary systems may employ molecular architectures fundamentally different from those used by contemporary terrestrial organisms [20–23,50–53].

3.1.2. Organic Complexity Beyond Earth

Recent asteroid-return missions have demonstrated that sophisticated organic chemistry occurs naturally in extraterrestrial environments. Samples from Bennu and Ryugu contain amino acids, nucleobases, sugars, ammonia-rich compounds, and complex macromolecular organic matter (MOM) [26–28]. These discoveries indicate that chemically diverse molecular inventories are not unique to Earth and may be widespread throughout planetary systems. The presence of such chemically rich environments expands the range of plausible molecular substrates available for prebiotic evolution. Although the biological significance of these compounds remains uncertain, they demonstrate that planetary chemistry can generate a remarkable diversity of molecular building blocks capable of participating in increasingly complex chemical systems [26–28]. Together, laboratory studies of alternative hereditary polymers and discoveries of complex extraterrestrial organic matter suggest that the chemical foundations of heredity may be substantially broader than those preserved in modern biology.

These observations motivate consideration of organizational frameworks capable of integrating diverse molecular components into persistent hereditary systems. The concentrations of amino acids, sugars, nucleobases, and related compounds detected in Bennu and Ryugu samples are generally low and do not necessarily imply that extraterrestrial delivery provided the dominant source of prebiotic substrates on the early Earth [26–28]. Rather, these discoveries demonstrate that abiotic synthesis of biologically relevant compounds occurs naturally in planetary environments [26–28]. The greater challenge remains identifying geochemical settings capable of producing, concentrating, and sustaining such molecules at levels sufficient to support prebiotic evolution before emerging biological systems consumed available resources.

3.1.3. Implications for Alternative Hereditary Systems

Collectively, these observations suggest that early hereditary systems may have explored a broader chemical landscape than that preserved in modern biology. The central challenge is therefore not simply to explain the origin of RNA, but to understand how hereditary processes emerge from interacting molecular populations. This perspective motivates consideration of organizational frameworks in which heredity is distributed across molecular ecosystems rather than confined to a single informational polymer.

3.2. Stable Molecular Networks

A major challenge in origins-of-life research is explaining how simple chemical mixtures transition into organized systems capable of persistence, information storage, and evolution. Modern microbial systems demonstrate that biological information can be distributed across interacting molecular components rather than residing solely within genomes, suggesting possible analogies for earlier stages of molecular evolution. Individual molecules are continually synthesized, modified, and degraded, yet complex biological systems maintain network stability despite continual molecular turnover. This observation

suggests that heredity may reside not only within individual molecules but also within the relationships among interacting molecular components.

Stable Molecular Networks (SMNs) are defined here as persistent interacting systems composed of informational polymers, catalytic oligomers, compartments, metabolites, and environmental feedback mechanisms that maintain organizational structure through time despite continual molecular turnover. SMNs are not proposed to be fully Darwinian systems. Rather, they represent molecular organizations capable of persistence, amplification, and limited hereditary continuity that may have preceded true Darwinian evolution. Within such systems, hereditary information is distributed across networks of interactions rather than residing exclusively within individual molecules (Box 2; Figure 2).

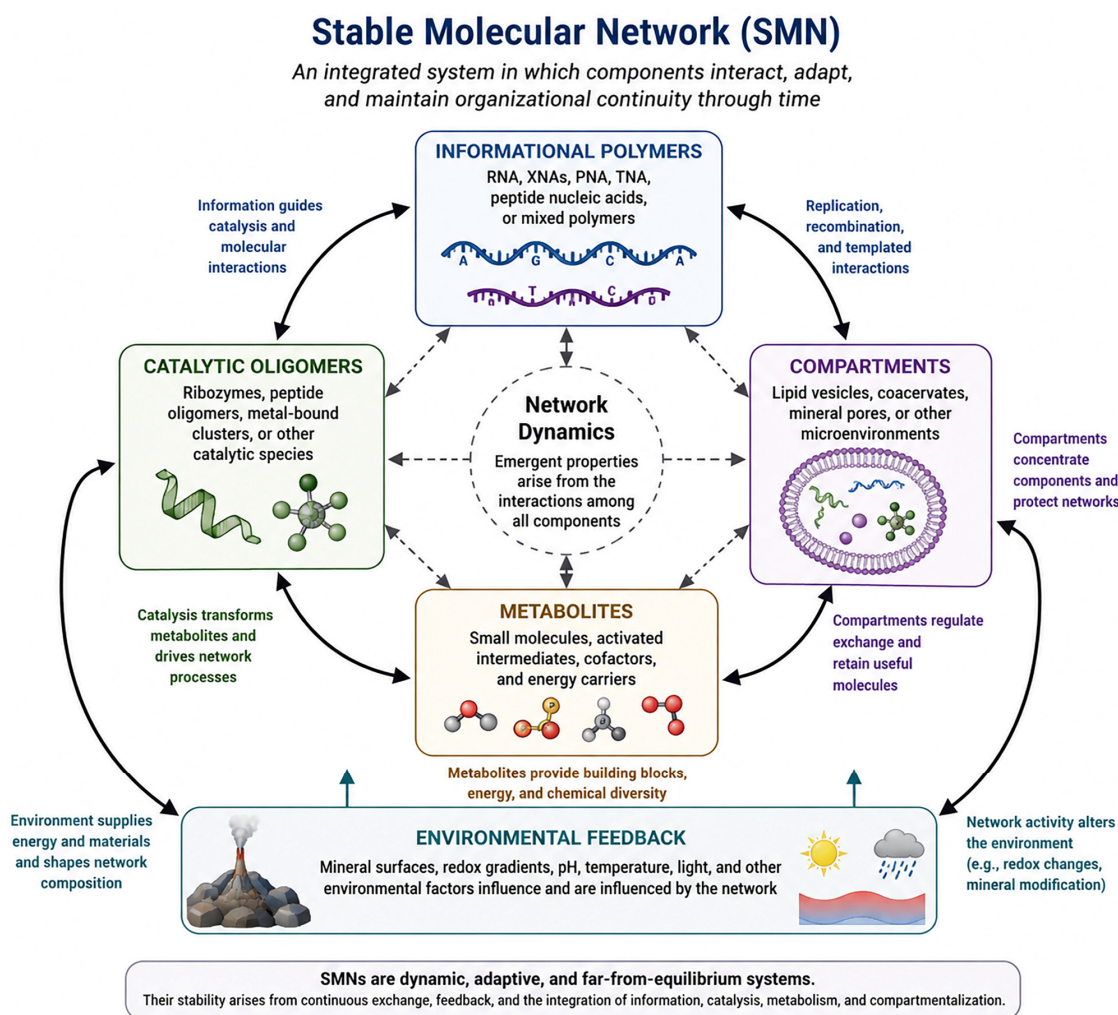


Figure 2. Stable Molecular Network (SMN). Proposed framework illustrating the major components of a Stable Molecular Network. SMNs consist of interacting informational polymers, catalytic oligomers, metabolites, compartments, and environmental feedback processes that collectively maintain organizational continuity despite continual molecular turnover. Informational polymers store and transmit information, catalytic oligomers drive chemical transformations, metabolites provide building blocks and energy, and compartments concentrate and regulate molecular interactions. Environmental conditions both influence and respond to network activity, creating reciprocal feedback between the system and its surroundings. The persistence of an SMN arises from the dynamic integration of these components through information exchange, catalysis, compartmentalization, metabolism, and environmental coupling. In this framework, heredity is viewed as an emergent property of the network rather than of any single molecular species.

Box 2. Definition.

Stable Molecular Network (SMN): A persistent interacting system of informational polymers, catalytic oligomers, compartments, and environmental feedback capable of maintaining organizational structure and propagating hereditary through time despite continual molecular turnover.

3.2.1. Compositional Inheritance

One route toward pre-cellular heredity involves compositional inheritance, in which the collective composition of molecular assemblies is transmitted through growth, division, or compartmentalization. Models such as the Graded Autocatalysis Replication Domain (GARD) framework demonstrate how molecular populations can exhibit heredity-like behavior without requiring genome-based replication [43,44]. Such systems illustrate that heredity can be distributed across molecular communities rather than encoded within a single informational polymer.

3.2.2. Autocatalytic Networks

Autocatalytic networks provide another example of distributed heredity. In these systems, interacting molecules collectively promote one another's formation, creating self-sustaining chemical organizations capable of persistence and evolution [54,55]. Theoretical and experimental studies suggest that autocatalytic networks may arise spontaneously under appropriate conditions and may represent important intermediates between prebiotic chemistry and biological evolution [54,55].

3.2.3. Compartmentalized Molecular Communities

Compartmentalization provides an important mechanism for maintaining molecular interactions. Vesicles, coacervates, mineral pores, and related structures can concentrate molecules, facilitate reaction networks, and promote localized evolutionary processes [56,57]. Rather than functioning as primitive cells, such compartments may initially have served as ecological habitats for interacting molecular populations. By increasing local concentrations and restricting diffusion, compartmentalization allows molecular interactions to persist long enough for selection, amplification, and diversification to occur. These properties make compartments a natural component of SMN. Comparable principles are observed in modern microbial biofilms, hydrothermal vent consortia [39–41], and other spatially structured microbial communities, where compartmentalization and local interactions influence evolutionary dynamics.

3.2.4. Stable Molecular Networks as Evolutionary Units

Taken together, compositional inheritance, autocatalytic networks, and compartmentalization suggest that evolutionary processes can occur at the level of interacting molecular communities. Within this framework, the fundamental unit of early evolution may have been neither the gene nor the organism, but the stable molecular network itself. Selection acts on the persistence and organizational properties of entire systems rather than individual molecules (Table 2).

Table 2. Representative Mechanisms Supporting Distributed Heredity. The table illustrates a progression from sequence-based replication to increasingly distributed forms of heredity in which system persistence emerges from molecular compositions, autocatalytic networks, compartmentalized communities, and molecular ecosystems. Within the framework proposed here, Stable Molecular Networks (SMNs) and Precellular Polymer Consortia (PPCs) represent hypothetical intermediates linking planetary chemistry to cellular life.

Mechanism	Primary Hereditary Carrier	Requires Genome-Based Replication?	Proposed Unit of Selection	Representative Examples
RNA Replication	RNA sequences	Yes	Replicating RNA molecules	RNA World; ribozymes [1–5]
Compositional Inheritance	Molecular compositions	No	Molecular assemblies	GARD model [43,44]
Autocatalytic Networks	Catalytic reaction networks	No	Chemical networks	RAF theory; autocatalytic sets [54,58]
Catalytic Oligomer Amplification	Short catalytic oligomers	Not necessarily	Interacting oligomer populations	Catalytic RNA networks [59,60]
Compartmentalized Molecular Communities	Molecular contents of compartments	No	Vesicle or coacervate populations	Protocells; fatty-acid vesicles [61–64]
Stable Molecular Networks (SMNs)	Distributed molecular interactions	No	Molecular ecosystems	Integrated polymer–compartment systems (this review)
Precellular Polymer Consortia (PPCs)	Compartments plus distributed molecular interactions	No	Molecular lineages	PPC hypothesis (this review)

3.3. Stable Molecular Networks as an Intermediate Stage Between Chemistry and Life

The observations summarized above motivate a broader view of heredity than that provided by canonical RNA-based models alone. Alternative informational polymers, Stable Molecular Networks (SMNs), compartmentalized molecular communities, and distributed hereditary systems collectively suggest that multiple pathways toward heredity may have existed during the earliest stages of planetary evolution. Modern microbial systems provide useful examples of distributed information processing, molecular memory, and ecological organization that may illuminate these transitions. Among these systems, CRISPR-Cas adaptive memory systems provide a particularly relevant microbial example of hereditary information storage and amplification [29–31], while eukaryotic piRNA pathways illustrate how molecular populations can participate in distributed information processing and molecular memory (Figure 3). The following section examines these contemporary systems as conceptual analogs for network behaviors that may have operated within pregenomic hereditary systems.

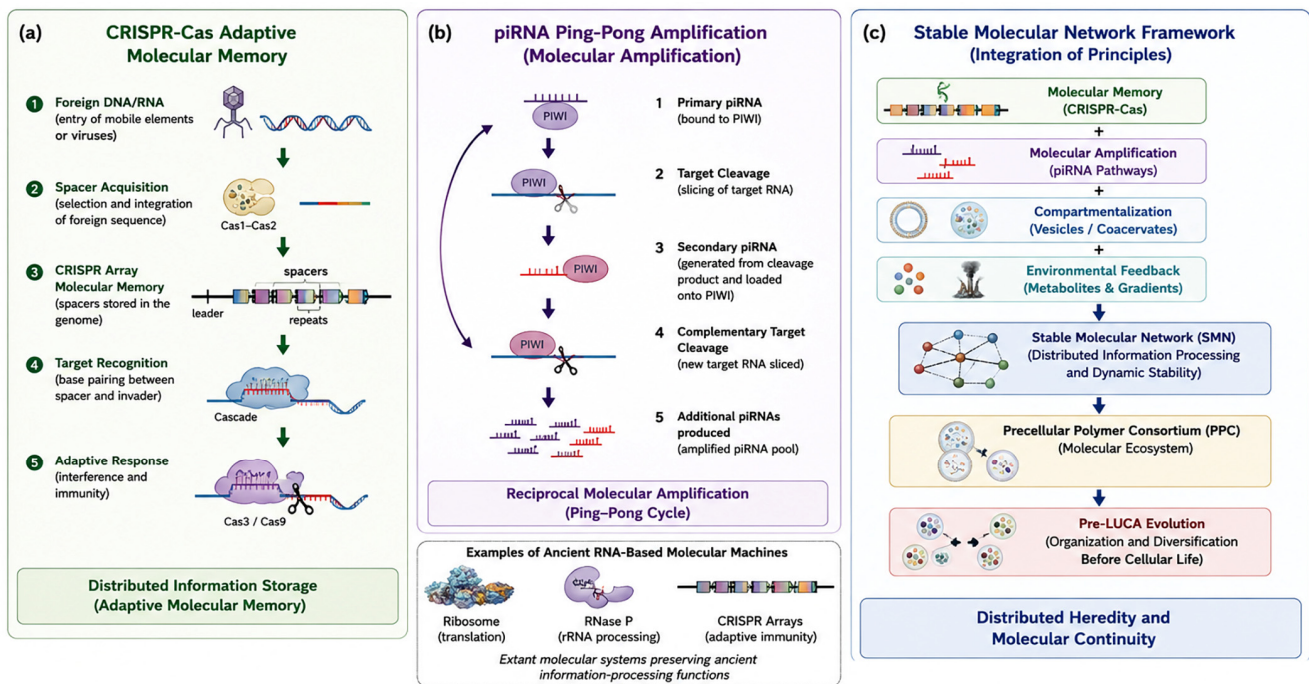


Figure 3. Contemporary biological analogues of Stable Molecular Networks (SMNs). (a) CRISPR-Cas systems in bacteria and archaea illustrate adaptive molecular memory through acquisition, storage, and reuse of foreign nucleic acid sequences. (b) piRNA pathways provide an example of reciprocal molecular amplification through the ping-pong cycle, in which interacting RNA populations generate self-reinforcing information propagation. (c) Conceptual integration of molecular memory, amplification, compartmentalization, and environmental feedback within the Stable Molecular Network framework. Although CRISPR-Cas and piRNA systems evolved within modern cellular organisms and are not proposed as direct relics of prebiotic evolution, they illustrate network behaviors that may have contributed to hereditary systems operating before LUCA.

4. Contemporary Molecular Memory and Amplification Systems as Conceptual Analogs of Stable Molecular Networks

The Stable Molecular Network (SMN) framework proposes that molecular continuity, amplification, and persistence emerged within interacting molecular systems before the appearance of modern genomes and cells. Because direct evidence from such early systems is unavailable, contemporary biological processes may provide useful conceptual analogs for understanding how distributed hereditary processes can operate. Modern molecular memory and amplification systems are highly derived biological innovations and are not proposed here as direct relics of prebiotic evolution. Nevertheless, they illustrate organizational principles that may be relevant to early molecular evolution.

Among these interacting molecular systems, CRISPR-Cas adaptive memory pathways in bacteria [29–31] and archaea and PIWI-interacting RNA (piRNA) pathways in eukaryotes are particularly informative because they combine information storage, molecular recognition, amplification, and persistence within distributed molecular networks [65–68]. The purpose of this section is therefore not to argue that CRISPR or piRNA systems are ancestral, but rather to examine how modern molecular memory systems demonstrate principles of molecular continuity that may also have characterized pregenomic hereditary systems operating within SMNs.

4.1. CRISPR-Cas Systems as Models of Molecular Memory

Adaptive Memory in Archaea and Bacteria

CRISPR-Cas systems provide adaptive immunity in bacteria and archaea through acquisition, storage, and reuse of foreign nucleic acid sequences [29–31]. Short fragments of invading DNA are incorporated into CRISPR arrays, generating molecular records of previous encounters that can subsequently guide recognition and destruction of related invaders [29–31]. Although CRISPR evolved within fully cellular organisms, it demonstrates that hereditary information can be stored, updated, and utilized within distributed molecular systems rather than residing solely within conventional genomes. These properties make CRISPR-Cas systems useful examples of molecular memory and information persistence relevant to the SMN framework.

4.2. piRNA Biology

4.2.1. Overview of piRNA Pathways

piRNAs are small noncoding RNAs that associate with PIWI proteins and function primarily in transposon regulation and maintenance of genome integrity [65–67]. From the perspective of the SMN framework, the importance of piRNAs lies not in their specific biological functions but in their demonstration that information can be stored, transmitted, and amplified through interacting populations of small RNAs. Their widespread conservation across animal lineages illustrates the evolutionary success of RNA-guided information processing and provides a useful conceptual example of distributed molecular memory operating within complex biological systems.

4.2.2. The Ping-Pong Amplification Cycle

One of the most distinctive features of piRNA biology is the ping-pong amplification cycle [65,67,69,70]. In this process, complementary RNA populations generate additional piRNAs through reciprocal cleavage reactions, creating a self-reinforcing amplification system. Importantly, amplification occurs through interactions among populations of molecules rather than through replication of a single informational template. Heredity therefore emerges from the dynamics of interacting molecular communities.

Although modern piRNA systems depend upon cellular machinery, the organizational principle of reciprocal amplification may have broader relevance. Molecular populations capable of recognizing, modifying, and regenerating one another could potentially amplify information without requiring modern genomes or chromosomes. Within the SMN framework, catalytic oligomers interacting with persistent polymers may have produced analogous amplification dynamics. While the chemistry would differ substantially from contemporary piRNA pathways, both systems illustrate how hereditary information can be propagated through interacting molecular populations rather than individual replicators (Table 3).

Together, CRISPR-Cas and piRNA systems demonstrate that biological continuity can arise through distributed molecular interactions involving memory, recognition, amplification, and inheritance. Although these systems evolved within fully cellular organisms, they illustrate emergent organizations that may have characterized hypothetical hereditary networks operating before genome-based cellular life.

Table 3. Contemporary molecular memory and amplification systems as conceptual analogs of Stable Molecular Networks. CRISPR-Cas systems provide examples of adaptive molecular memory in bacteria and archaea, whereas piRNA pathways illustrate reciprocal molecular amplification and distributed information processing. Neither system is proposed as a direct relic of prebiotic evolution. Rather, they demonstrate organizational principles—including memory, recognition, amplification, and compartment-mediated information transfer—that may have characterized hypothetical hereditary systems operating before genome-based cellular life.

Modern System	Biological Function	Conceptual Analog in SMNs	Relevance to Early Heredity
CRISPR arrays	Storage of acquired sequence information	Persistent molecular memory	Maintenance of hereditary information
CRISPR-Cas complexes	Recognition and processing of foreign nucleic acids	Molecular recognition systems	Information-guided selection and response
piRNA populations	Sequence-specific regulation	Informational polymers	Recognition and transmission of hereditary information
Ping-pong amplification	Reciprocal generation of piRNAs	Molecular amplification networks	Population-level information amplification
Vesicle-mediated RNA transfer	Movement of information between compartments	Exchange among molecular communities	Hereditary transfer without cellular reproduction
Distributed regulatory networks	Collective information processing	Stable Molecular Networks (SMNs)	Distributed heredity across molecular populations

4.3. Vesicles and Compartmentalized Information Transfer

4.3.1. Vesicle-Mediated Transport

Modern organisms employ extracellular vesicles, exosomes, and related structures to transport RNAs, proteins, metabolites, and signaling molecules between cells [61–64]. These systems demonstrate that hereditary components can move between compartments independently of cellular reproduction. More generally, they illustrate how compartmentalization can concentrate, protect, and exchange molecular information among interacting populations.

4.3.2. From Vesicles to Molecular Ecosystems

Compartmentalization is also a central component of the SMN framework. Vesicles, coacervates, mineral pores, and related structures create localized environments that concentrate polymers, catalytic oligomers, metabolites, and cofactors [61–64]. These compartments need not possess genomes or metabolism in the modern sense to participate in evolutionary processes. Rather, they may function as components of larger molecular ecosystems in which continuity of heredity emerges from interactions among compartmentalized molecular populations. The concepts of molecular memory, amplification, distributed heredity, and compartmentalized exchange provide a foundation for considering how molecular ecosystems could diversify into distinct lineages. The next section explores this possibility through the framework of Precellular Polymer Consortia (PPCs) and precellular speciation.

5. Precellular Polymer Consortia and Precellular Speciation

5.1. From Stable Molecular Networks to Molecular Ecosystems

5.1.1. Emergence of Molecular Ecosystems

Stable Molecular Networks (SMNs) provide a conceptual framework for understanding how interacting informational polymers, catalytic oligomers, metabolites, and environmental feedback systems may generate persistent hereditary organization before the emergence of modern cells. As these interactions become increasingly interconnected,

collections of SMNs may form larger molecular ecosystems capable of maintaining continuity through time. In such systems, persistence arises not from replication of individual molecules alone, but from the collective behavior of interacting molecular populations.

Modern microbial communities illustrate how system-level properties can emerge from distributed interactions among diverse components. Although microbial ecosystems are vastly more complex than hypothetical prebiotic systems, they demonstrate that continuity, adaptation, and information processing can arise from interactions among many participants rather than isolated replicators. These observations motivate consideration of molecular ecosystems as potential intermediates between planetary chemistry and cellular life.

5.1.2. Compartmentalization and Organizational Continuity

Compartmentalization is expected to play a critical role in maintaining molecular ecosystems. Vesicles, coacervates, mineral pores, and related structures can concentrate polymers, catalytic oligomers, metabolites, and cofactors while simultaneously restricting diffusion. Such compartments increase opportunities for repeated interactions among molecular populations and may promote the persistence of successful organizational states. Rather than viewing compartments solely as passive containers, the SMN framework treats them as active participants in hereditary processes. Compartments influence which molecular interactions occur, which components are retained, and how information is exchanged among neighboring systems. Organizational continuity therefore emerges from interactions among molecules, compartments, and environmental conditions.

5.1.3. Coacervates and Membrane Compartments

A central question in origins-of-life research concerns the nature of the compartments that first concentrated informational polymers and catalytic molecules [71–74]. Two major classes of compartments have received particular attention: membraneless condensates such as coacervates and membrane-bound vesicles [73,74]. Both provide mechanisms for concentrating dilute molecular populations and promoting interactions that would otherwise occur inefficiently in bulk solution.

Coacervates are liquid–liquid phase-separated droplets formed through interactions among charged polymers, proteins, nucleic acids, and other macromolecules [73,74]. Experimental studies have demonstrated that coacervates can concentrate RNA, peptides, metabolites, and catalytic molecules by several orders of magnitude relative to their surrounding environment [75–77]. In addition, both proteins and nucleic acids often exhibit enhanced folding, stability, and catalytic activity within coacervate environments owing to increased local concentrations and altered physicochemical conditions [75–77]. These observations suggest that coacervates may have provided favorable settings for early molecular evolution and have therefore been proposed as plausible precellular compartments.

Despite these attractive properties, coacervates have not yet been shown to support long-term hereditary continuity or open-ended evolution. While they can concentrate molecules and facilitate chemical reactions, the extent to which they can maintain persistent hereditary states over many generations remains uncertain [75–77]. At present, there is no experimental demonstration that coacervate populations alone can sustain stable inheritance, ecological diversification, or lineage formation comparable to biological evolution.

Membrane-bound compartments offer a complementary set of advantages. Lipid vesicles and related membrane structures provide physical boundaries that protect internal contents while permitting selective exchange with the external environment [78]. Membranes can move, fuse, divide, and transport molecular cargo between compartments [78]. These properties enable interactions among compartmentalized systems while simulta-

neously preserving organizational identity [78]. Membrane fusion and vesicle-mediated exchange also provide potential mechanisms for horizontal transfer of informational polymers, catalytic oligomers, and metabolites [79].

From the perspective of the PPC framework, membranes may be particularly important because they facilitate ecological interactions among molecular ecosystems. Mobile compartments can colonize new environments, exchange molecular information, respond to environmental stress, and maintain system persistence despite fluctuating conditions. Partial isolation among membrane-bound systems may also promote divergence, allowing distinct organizational states to emerge and persist. Such processes provide plausible pathways toward the precellular speciation mechanisms discussed below.

The PPC model does not exclude a role for coacervates. Indeed, coacervates may have acted as important concentrators of polymers and catalysts within early molecular ecosystems. However, we propose that membrane-containing compartments ultimately provided a more robust substrate for long-term hereditary continuity, ecological diversification, and evolutionary innovation.

Future experimental studies comparing coacervate-based and membrane-based molecular communities may help clarify the relative contributions of these compartment types to the emergence of early life. Importantly, coacervates may have functioned as concentrators within larger membrane-containing molecular ecosystems rather than representing mutually exclusive alternatives.

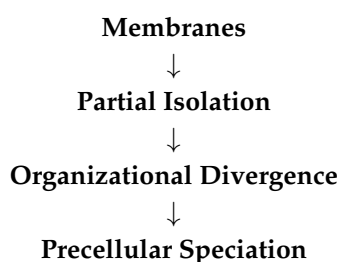
5.1.4. Why Membranes May Have Enabled Precellular Speciation

While both coacervates and membrane-bound compartments can concentrate polymers and catalytic molecules, membrane systems possess properties that may be particularly important for evolutionary diversification. Membrane vesicles can move through their environment, fuse with other compartments, exchange molecular cargo, and maintain distinct internal compositions over extended periods. These properties allow information to flow among molecular ecosystems while preserving organizational identity.

Mobility would enable compartments to encounter new environments and molecular populations, increasing opportunities for innovation. Fusion and vesicle-mediated exchange could generate novel combinations of informational polymers, catalytic oligomers, and metabolites. At the same time, compartment boundaries would limit complete mixing, allowing distinct organizational states to persist. These exchanges may represent pregenomic analogues of horizontal gene transfer [80,81], allowing innovations arising within one molecular ecosystem to spread through larger PPC communities.

This balance between information exchange and partial isolation may have promoted ecological diversification among molecular ecosystems. Compartments occupying different environments, exhibiting different fusion compatibilities, or maintaining different molecular compositions could gradually diverge over time. As organizational differences accumulated, molecular exchange would become less effective, reinforcing divergence.

We therefore propose that membranes may have facilitated precellular diversification by balancing information exchange with partial isolation, allowing molecular ecosystems to diverge into increasingly distinct organizational states.



Although speculative, this framework generates experimentally testable predictions regarding the relative abilities of membrane-bound and coacervate-based systems to maintain organizational continuity, exchange molecular information, and diversify under ecological selection.

5.2. Precellular Polymer Consortia

5.2.1. Definition of PPCs

We define Precellular Polymer Consortia (PPCs) as compartmentalized molecular ecosystems composed of interacting informational polymers, catalytic oligomers, metabolites, and environmental feedback systems that collectively maintain organizational continuity. PPCs are not proposed to possess genomes, chromosomes, or biological reproduction in the modern sense. Instead, continuity arises from persistence of network organization. Within PPCs, hereditary information is distributed across interacting molecular populations. The state of the system depends not only upon the presence of individual molecules but also upon their relationships, relative abundances, spatial organization, and interaction histories. Heredity therefore becomes a property of the consortium rather than any single molecular species (Figure 4).

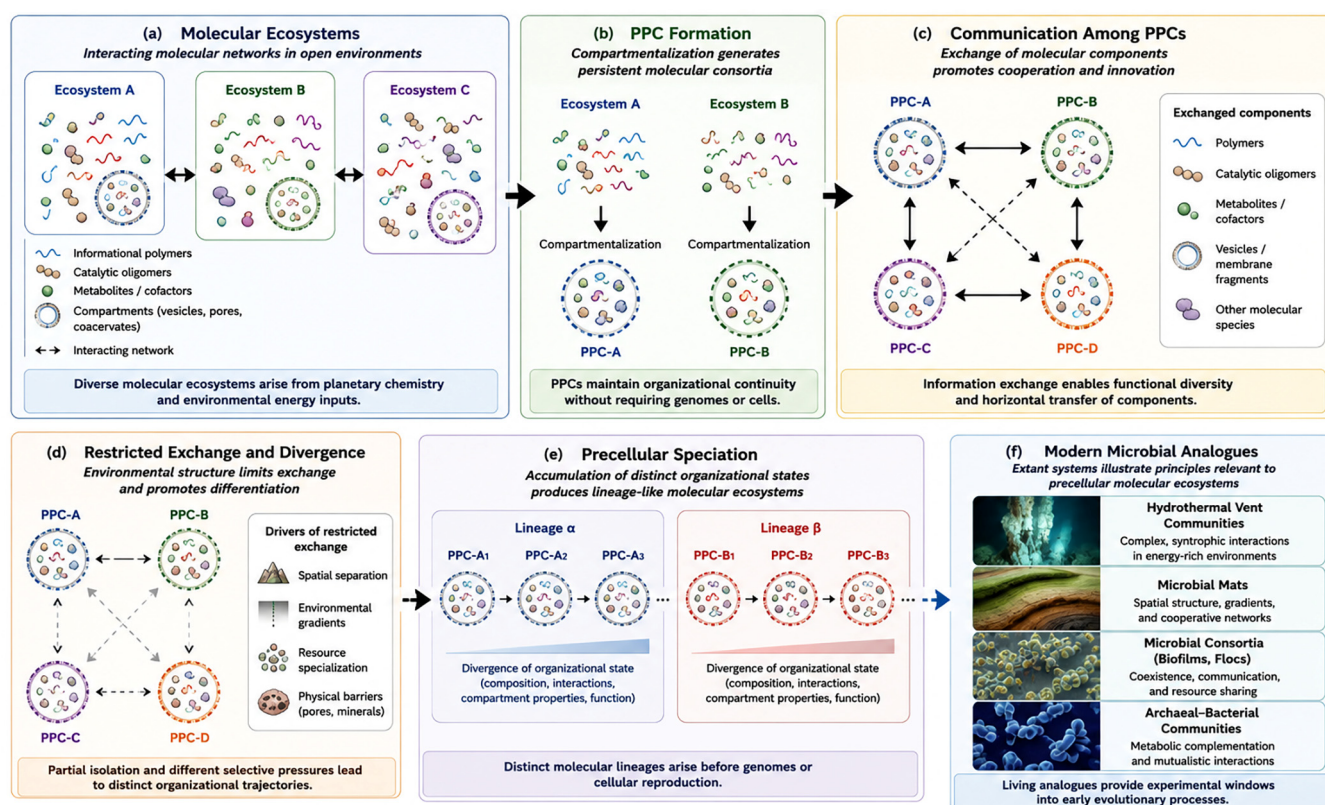


Figure 4. Proposed diversification of molecular ecosystems into Precellular Polymer Consortia (PPCs) and precellular lineages. (a) Interacting molecular ecosystems composed of informational polymers, catalytic oligomers, metabolites, and compartments. (b) Compartmentalization generates PPCs capable of maintaining organizational continuity. (c) Exchange of molecular components among PPCs promotes communication and innovation. (d) Restricted exchange resulting from environmental gradients, spatial separation, or resource specialization promotes divergence. (e) Accumulation of distinct organizational states leads to precellular speciation and the emergence of lineage-like molecular ecosystems. (f) Modern microbial consortia, hydrothermal vent communities, and archaeal-bacterial ecosystems are proposed as living analogues for investigating organizational principles relevant to early evolution before LUCA.

5.2.2. PPCs as Evolutionary Units

PPCs provide a potential intermediate level of biological organization between SMNs and fully cellular life. Because PPCs consist of multiple interacting components, selection may operate on organizational states rather than individual replicators. Consortia that more effectively maintain information, utilize resources, or resist environmental perturbation would be expected to persist longer and contribute disproportionately to future molecular ecosystems. A useful analogy may be found in modern microbial consortia, where community-level interactions influence survival, metabolism, and ecological success [82]. While PPCs are hypothetical pre-cellular systems, both PPCs and microbial communities demonstrate that evolutionary processes can operate on interacting networks rather than isolated individuals.

5.3. Molecular Communication and Information Exchange

5.3.1. Exchange Among Molecular Ecosystems

For evolutionary diversification to occur, molecular ecosystems must be capable of exchanging information. Such exchange may occur through movement of polymers, catalytic oligomers, metabolites, vesicles, or other compartment-associated components [83,84]. These interactions can generate cooperative, competitive, or parasitic relationships among neighboring PPCs. Information exchange also creates opportunities for innovation. Novel molecular combinations arising in one consortium may spread to others, generating forms of horizontal transfer analogous to processes observed in modern microbial systems.

5.3.2. Restricted Exchange and Divergence

At the same time, complete mixing would prevent diversification. Partial isolation resulting from spatial separation, compartment boundaries, environmental gradients, or resource specialization may reduce exchange among PPCs. Such restrictions create opportunities for molecular ecosystems to follow distinct evolutionary trajectories. Modern microbial communities again provide useful analogies. Geographic isolation, ecological specialization, and limited gene flow contribute to diversification among microbial populations [83,84]. Similar principles may have operated in pre-cellular molecular ecosystems, although through entirely different molecular mechanisms.

5.4. Precellular Speciation

5.4.1. Organizational Divergence

The concept of precellular speciation proposes that distinct molecular lineages could emerge before the appearance of genomes, chromosomes, and biological reproduction. Divergence would occur when PPCs accumulate different organizational states that become increasingly stable and self-reinforcing through time. Under this framework, lineage identity is defined by system persistence rather than genetic sequence. Molecular ecosystems possessing distinct combinations of polymers, catalytic activities, compartments, and interaction networks may therefore represent different evolutionary lineages.

A loose conceptual analogy may be drawn to the green beard effect proposed by Dawkins and subsequently formalized in evolutionary theory [85,86]. In biological systems, green beard mechanisms involve a recognizable trait, recognition of that trait in others, and preferential cooperation among individuals sharing the trait [85,86]. Although PPCs lack genes, organisms, and reproduction in the conventional sense, molecular ecosystems exhibiting preferential fusion, molecular exchange, or cooperative interactions with compositionally similar compartments may display analogous forms of assortative interaction. Such compatibility-based exchange could reinforce organizational differences among molecular ecosystems and thereby contribute to divergence and precellular speciation.

5.4.2. Ecological Selection Among PPCs

Environmental conditions are expected to influence which PPCs persist. Differences in energy availability, resource utilization, stability, or resistance to perturbation may favor some organizational states over others. Selection therefore acts upon the performance of the consortium. This perspective is conceptually related to earlier proposals emphasizing communal evolution, network-based inheritance, and major evolutionary transitions in biological organization. PPCs extend these ideas by proposing that ecological diversification and lineage formation may have begun before modern cells emerged (Table 4).

Table 4. Comparison between biological speciation and the proposed process of precellular speciation. Although the mechanisms differ fundamentally, both processes involve progressive restriction of information exchange, divergence of interacting populations, ecological differentiation, and the emergence of increasingly distinct lineages. The proposed analogies are intended as conceptual rather than literal evolutionary equivalences.

Biological Evolution	Proposed Precellular Analogue
Reproductive isolation	Restricted information exchange
Mate recognition	Selective vesicle fusion or compartment compatibility
Genetic incompatibility	Molecular incompatibility
Gene flow	Molecular information flow
Ecological specialization	Compartment specialization
Population	Molecular ecosystem
Species	Molecular lineage
Natural selection	Selection on organizational states
Horizontal gene transfer	Exchange of polymers, metabolites, or vesicles
Evolution of populations	Evolution of molecular ecosystems

5.5. Implications for the Transition to Cellular Life

The PPC framework suggests that important evolutionary processes—including molecular memory, information exchange, ecological interactions, and selection—may have preceded modern genome-based reproduction. Rather than appearing suddenly with the first cells, these processes may have emerged gradually within increasingly organized molecular ecosystems. Extant microbial communities are not direct descendants of PPCs, nor are they primitive relics of pre-cellular evolution. However, microbial consortia, hydrothermal vent ecosystems [39–41], and archaeal-bacterial communities demonstrate how complex adaptive behavior can arise from distributed interactions among many components. These systems therefore provide useful experimental models for exploring organizational principles that may have contributed to the emergence of cellular life and the evolutionary transition leading to LUCA.

Although PPCs are hypothetical, several organizational features proposed for these systems—including distributed information processing, cooperative interactions, ecological specialization, and adaptive persistence—are observed in extant microbial communities. Modern archaea and bacteria therefore provide valuable experimental systems for investigating principles that may have operated during earlier stages of biological evolution (Box 3).

Box 3. Testable Predictions of the Stable Molecular Network and Precellular Polymer Consortia Frameworks.

A useful hypothesis should generate experimentally testable predictions. Although the Stable Molecular Network (SMN) and Precellular Polymer Consortia (PPC) frameworks are speculative, they suggest several observations that could be evaluated experimentally using synthetic molecular systems, laboratory evolution, microbial ecology, and comparative genomics.

- *Prediction 1. Membrane-bound molecular ecosystems should exhibit greater long-term organizational continuity than coacervate-only systems.* If membranes facilitate information retention, compartmentalization, and selective exchange, membrane-containing molecular communities should maintain stable organizational states for longer periods than equivalent coacervate-based systems [87].
- *Prediction 2. Partial isolation should promote diversification among molecular ecosystems.* Experimental populations of compartmentalized molecular systems subjected to limited exchange are predicted to diverge more readily than populations experiencing unrestricted mixing. Such divergence may represent an analogue of precellular speciation [83,84].
- *Prediction 3. Distributed catalytic networks should outperform isolated replicators under fluctuating environmental conditions.* Networks composed of interacting polymers, catalysts, and metabolites may exhibit greater robustness and adaptability than systems dependent upon a single dominant replicator, particularly under changing environmental conditions [63].
- *Prediction 4. Molecular exchange may accelerate innovation while preserving organizational identity.* Intermediate levels of fusion, vesicle exchange, or molecular transfer are predicted to generate novel combinations of functional molecules without eliminating differences among molecular ecosystems. Both excessive isolation and unrestricted mixing are expected to reduce diversification [83,84].
- *Prediction 5. CRISPR arrays may contain hierarchical signatures of molecular accretion.* Comparative analyses of archaeal and bacterial CRISPR systems may reveal structural or organizational patterns analogous to those identified in ribosomal RNA and RNase P accretion studies [34,88,89]. Such findings would support the broader concept that molecular memory systems can preserve historical information over evolutionary timescales.
- *Prediction 6. Community-level organization may emerge before genome-level optimization.* Synthetic molecular ecosystems may exhibit selection on network properties, resource utilization, stability, or information exchange before the appearance of highly efficient individual replicators. This would support the hypothesis that system persistence can precede modern genome-based inheritance [90].
- *Prediction 7. Hydrothermal vent ecosystems may exhibit network behaviors relevant to PPCs.* Modern vent-associated microbial communities are predicted to provide experimentally accessible examples of distributed metabolism, ecological cooperation, compartmentalization, and information exchange that can be used to evaluate aspects of the PPC framework [39–41].

Collectively, these predictions shift the SMN/PPC framework from a purely conceptual model toward an experimentally tractable research program linking origins-of-life studies, microbial ecology, molecular evolution, and astrobiology.

6. Extant Microbial Systems as Windows into Early Evolution

6.1. Why Extant Microbial Systems Matter

The earliest stages of biological evolution cannot be observed directly because no confirmed fossils, genomes, or molecular records exist from the period preceding the Last Universal Common Ancestor (LUCA). Consequently, origins-of-life research relies primarily on three complementary approaches: laboratory studies of prebiotic chemistry, theoretical models of early hereditary systems, and comparative analyses of extant biological systems. While modern organisms are separated from the origin of life by billions of years of evolution, they retain molecular features and network behaviors that may provide insights into ancient evolutionary processes.

A common misconception is that contemporary archaea and bacteria represent primitive relics of early life. All extant organisms are highly evolved and have undergone extensive adaptation since the emergence of LUCA. Modern thermophilic archaea, chemolithotrophic bacteria, and hydrothermal vent microorganisms should therefore not be viewed as direct descendants of precellular systems. Rather, they serve as experimen-

tally accessible models for investigating biological principles that may have contributed to early evolution, including molecular memory, catalytic cooperation, compartmentalization, ecological interactions, and distributed information processing.

Several molecular systems provide particularly valuable examples of ancient biological organization. The ribosome and RNase P preserve catalytic RNA components that likely originated before the diversification of modern cellular lineages and may retain structural signatures of early molecular evolution [34,88,89]. CRISPR-Cas systems demonstrate how biological systems can acquire, store, and utilize molecular information over time through adaptive memory mechanisms. Although these systems evolved long after the origin of life, they illustrate general principles by which information can be accumulated, maintained, and transmitted within complex molecular networks.

At larger scales, microbial ecosystems reveal how biological organization can emerge from interactions among multiple participants. Hydrothermal vent communities, microbial mats, and syntrophic consortia exhibit distributed metabolism, resource sharing, ecological specialization, and cooperative interactions that extend beyond individual organisms. These systems demonstrate that adaptive behavior and evolutionary persistence can arise from networks of interacting components rather than isolated entities. Such organizational properties are conceptually relevant to Stable Molecular Networks (SMNs) and Precellular Polymer Consortia (PPCs), which similarly emphasize interactions among multiple molecular participants.

The goal of this section is therefore not to identify living representatives of precellular evolution. Instead, we examine extant molecular systems and microbial communities as analogues that may illuminate principles relevant to early hereditary networks. By studying how modern organisms store information, coordinate metabolism, maintain organizational continuity, and interact within complex ecological systems, we may gain insight into mechanisms that helped bridge the transition from planetary chemistry to cellular life.

6.2. Extant Molecular Fossils of Early Evolution

Although the earliest hereditary systems are no longer accessible directly, several ancient molecular systems preserve structural and functional features that may illuminate early evolutionary transitions. Among the most informative are the ribosome, RNase P, and CRISPR-Cas pathways. These systems differ substantially in age, function, and evolutionary history, yet all demonstrate mechanisms by which information can be stored, processed, and maintained within complex molecular networks. As such, they provide useful models for exploring organizational principles relevant to Stable Molecular Networks (SMNs) and Precellular Polymer Consortia (PPCs).

6.2.1. Ribosomal Accretion and Ancient RNA Structures

The ribosome is among the most ancient and universally conserved molecular machines in biology. Comparative structural analyses suggest that modern ribosomes evolved through a process of accretion in which new RNA and protein components were progressively added to simpler ancestral cores [34,88,89]. These studies indicate that the ribosomal peptidyl transferase center is composed primarily of RNA and likely predates many associated protein components, supporting the hypothesis that catalytic RNAs played central roles during early evolution.

Accretion models provide an important conceptual framework for origins-of-life research because they demonstrate how complex molecular systems can arise through incremental addition and integration of functional modules [34,88,89]. Rather than emerging fully formed, biological complexity may develop through successive layers of organization

that retain traces of earlier evolutionary stages. The ribosome therefore serves not only as a molecular fossil of ancient biology but also as an example of how informational and catalytic functions can become integrated within stable molecular networks [34,88,89].

6.2.2. RNase P and the Persistence of Catalytic RNA

RNase P provides a second example of an ancient RNA-based molecular system. In many organisms, RNase P retains a catalytic RNA component capable of processing precursor transfer RNAs, demonstrating that RNA-catalyzed reactions remain essential in modern biology [34,88,89]. The widespread conservation of RNase P across all domains of life suggests deep evolutionary origins and supports the broader view that catalytic RNAs were likely more prevalent during earlier stages of biological evolution.

Like the ribosome, RNase P illustrates how functional information can be distributed across interacting molecular components rather than residing exclusively within protein enzymes [34,88,89]. The persistence of catalytic RNA within contemporary organisms provides direct evidence that RNA-based functionality remains compatible with highly evolved biological systems and may represent a surviving remnant of more extensive RNA-centered networks that existed before LUCA.

6.2.3. CRISPR-Cas Systems and Molecular Memory

CRISPR-Cas systems provide a distinct perspective on early evolution by demonstrating how biological systems can acquire, store, and utilize molecular information over time. Through the incorporation of foreign nucleic acid sequences into CRISPR arrays, bacteria and archaea generate adaptive molecular records of previous environmental encounters. These stored sequences can subsequently guide sequence-specific recognition and defense responses, creating a form of molecular memory that persists across generations.

Although CRISPR-Cas systems evolved long after the origin of life, they illustrate general principles of information storage, retrieval, and amplification within distributed molecular networks. Particularly noteworthy is the prevalence of CRISPR systems among archaeal lineages [91,92], suggesting that adaptive molecular memory has been an important feature of microbial evolution for extended periods of Earth history.

An intriguing question is whether CRISPR arrays preserve deeper evolutionary signatures beyond their immediate immunological functions. Like accretion models proposed for ribosomes and RNase P, future studies may determine whether CRISPR loci contain hierarchical patterns of growth, structural conservation, or historical layering that reflect broader principles of molecular evolution. Regardless of the outcome, CRISPR systems demonstrate that information can be accumulated and maintained within complex molecular architectures, providing a modern example of system persistence that is conceptually relevant to the SMN and PPC frameworks.

Collectively, the ribosome, RNase P [33,34,88,93], and CRISPR-Cas systems illustrate three complementary aspects of biological organization: the integration of catalytic and informational functions, the persistence of ancient RNA-based activities, and the storage of adaptive molecular information. While none of these systems directly represent precellular evolution, they provide experimentally accessible examples of network behaviors that may have contributed to the emergence of heredity before LUCA.

An unresolved question is whether CRISPR arrays preserve deeper evolutionary signatures analogous to those identified through ribosomal accretion analyses. Ribosomal RNA and RNase P retain structural evidence of hierarchical growth and evolutionary layering over billions of years [33,34,88,93]. Whether similar approaches can be applied to CRISPR loci remains unknown. If patterns of spacer acquisition, repeat evolution, or array architecture preserve detectable historical signatures, CRISPR systems may provide a new

class of molecular fossils capable of illuminating the evolution of biological information storage itself (Box 4).

Box 4. Can CRISPR arrays preserve deep evolutionary history?

The ribosome and RNase P have been described as molecular fossils because their structures retain signatures of evolutionary accretion that can be reconstructed through comparative analysis [33,34,88,93]. These studies suggest that complex molecular systems often arise through the gradual addition of functional modules, leaving detectable traces of earlier evolutionary states. Whether CRISPR-Cas systems preserve analogous signatures remains unknown. CRISPR arrays accumulate spacer sequences through repeated acquisition events and therefore contain chronological records of past interactions with mobile genetic elements. This raises the possibility that CRISPR loci may preserve hierarchical patterns of growth, organization, and diversification that extend beyond their immediate immunological functions. Several questions emerge from this perspective:

- Do CRISPR arrays exhibit structural or organizational features consistent with evolutionary accretion?
- Can spacer architectures preserve long-term signatures of historical growth and diversification?
- Are some CRISPR repeat structures more ancient than others, analogous to conserved ribosomal cores?
- Can comparative analyses reconstruct the evolutionary history of molecular memory systems in a manner like ribosomal evolutionary archaeology?
- Could CRISPR loci represent a distinct class of molecular fossils that illuminate the origins and evolution of biological information storage?

Although entirely speculative, these questions suggest a potential research direction linking microbial genomics, evolutionary biology, and origins-of-life studies. If CRISPR arrays retain detectable signatures of deep evolutionary history, they may provide a complementary framework to ribosomal accretion analyses for investigating how information storage systems evolve through time.

6.3. Extant Microbial Ecosystems as Analogues of Precellular Polymer Consortia

The Precellular Polymer Consortia (PPC) framework proposes that early hereditary systems may have consisted of interacting molecular ecosystems rather than isolated replicators. Although direct evidence for such systems is unlikely to survive, modern microbial communities provide experimentally accessible examples of how distributed interactions among multiple participants can generate emergent organizational properties [94]. These systems are not proposed as direct descendants of precellular molecular ecosystems. Rather, they illustrate biological principles—including compartmentalization, cooperation, resource exchange, ecological specialization, and distributed information processing—that may be relevant to understanding evolutionary transitions preceding LUCA.

6.3.1. Hydrothermal Vent Archaea as Models of Early Ecological Organization

Hydrothermal vent environments remain among the most extensively studied settings for the origin and early evolution of life because they provide continuous energy fluxes, steep geochemical gradients, mineral catalysts, and naturally compartmentalized microenvironments [39–41]. Modern vent-associated microorganisms therefore offer valuable opportunities to examine how biological organization can emerge under environmentally driven conditions.

Many archaeal lineages inhabiting hydrothermal systems possess physiological characteristics that are particularly relevant to origins-of-life research. Hyperthermophilic genera such as *Methanopyrus*, *Pyrococcus*, and *Thermococcus* thrive in environments characterized by high temperatures, strong redox gradients, and chemically dynamic conditions [95,96]. Other archaea, including *Ignicoccus hospitalis*, exhibit unusual membrane architectures and ecological interactions that highlight the diversity of organizational strategies present within contemporary microbial systems [97,98].

These organisms should not be viewed as primitive survivors of pre-cellular evolution. Rather, they are highly evolved microorganisms that have adapted to extreme environments over billions of years. Nevertheless, their dependence on chemolithotrophic energy sources, close coupling to geochemical processes, and participation in complex microbial communities provide useful models for exploring how metabolism, compartmentalization, and ecological organization may become integrated within evolving systems.

Hydrothermal vent ecosystems are also relevant because they provide continuous geochemical energy sources capable of supporting protometabolic processes. Hydrogen, carbon dioxide, sulfur compounds, iron minerals, and natural redox gradients can drive carbon fixation and energy transduction independently of sunlight [87]. Many origins-of-life models therefore view vent systems not simply as habitats for microorganisms but as environments in which metabolism-like processes may have emerged before modern cellular life.

The discovery of Asgard archaea has further emphasized the importance of archaeal evolution for understanding early biological complexity. These lineages encode numerous eukaryotic signature proteins and have reshaped views regarding the evolutionary relationship between archaea and eukaryotes [99]. Although Asgard archaea are highly derived modern organisms, their study illustrates how extant microbial systems can provide insights into major evolutionary transitions.

6.3.2. Syntrophic Consortia and Distributed Metabolism

Some of the clearest examples of distributed biological organization occur within syntrophic microbial consortia. In these systems, multiple species cooperate metabolically in ways that allow neither partner to function efficiently in isolation. Through the exchange of metabolites, reducing equivalents, and energy-rich intermediates, syntrophic communities achieve biochemical transformations that exceed the capabilities of individual organisms [100,101].

Particularly informative examples include associations between anaerobic methane-oxidizing archaea (ANME) [102,103] and sulfate-reducing bacteria [104,105]. These consortia exhibit tightly coordinated metabolic interactions in which resources, electrons, and biochemical functions are distributed among community members. Recent evidence for direct interspecies electron transfer further demonstrates that physiological integration can occur across multiple organisms, creating functional systems that operate at the community level rather than solely at the level of individual cells [106].

Although syntrophic communities have fully evolved biological systems, they illustrate an important principle underlying the PPC framework: adaptive organization can emerge from networks of interacting participants. Distributed metabolism, cooperative resource utilization, and collective responses to environmental conditions provide experimentally tractable examples of how system-level properties arise from interactions among many components.

6.3.3. Community-Level Organization and Ecological Cooperation

Microbial ecosystems frequently exhibit organizational properties that extend beyond individual organisms. Microbial mats, hydrothermal vent communities, archaeal-bacterial consortia, and other complex assemblages demonstrate how information processing, resource allocation, ecological specialization, and environmental adaptation can emerge through interactions among multiple participants [107]. In such systems, survival and persistence often depend as much on community structure as on the characteristics of individual species.

From the perspective of the PPC framework, these observations are significant because they demonstrate that biological organization can exist at multiple hierarchical levels simultaneously. Genes operate within genomes, genomes operate within cells, and cells operate within ecological networks. The resulting system-level properties frequently cannot be predicted solely from the behavior of individual components.

The relevance of these microbial ecosystems to origins-of-life research lies not in any direct evolutionary continuity, but in the organizational principles they illustrate. Distributed function, ecological cooperation, compartmentalization, resource exchange, and adaptive network behavior are all characteristics of modern microbial communities that may provide insight into how increasingly complex hereditary systems emerged before LUCA. While the molecular mechanisms would have differed substantially from those operating in contemporary organisms, extant microbial ecosystems demonstrate that interactions among many participants can generate stable and adaptive organizational states.

Collectively, hydrothermal vent archaea, syntrophic consortia, and complex microbial communities provide experimentally accessible models for investigating how distributed interactions contribute to persistence, adaptation, and evolutionary innovation. These properties make them valuable analogues for exploring the types of organizational processes envisioned within Stable Molecular Networks and Precellular Polymer Consortia (Figure 5; Table 5).

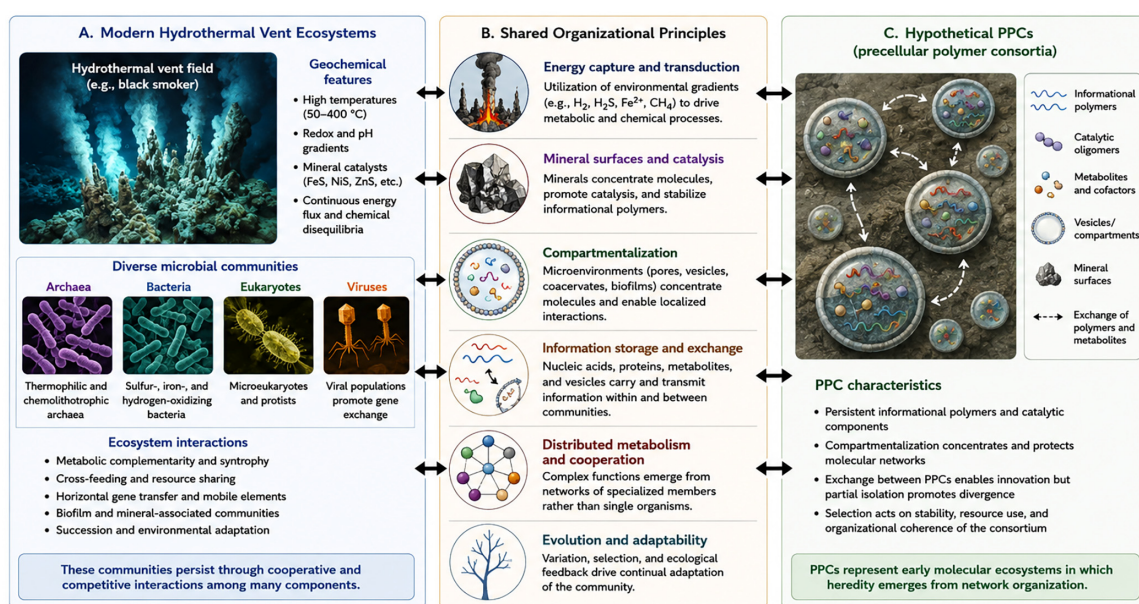


Figure 5. Hydrothermal vent ecosystems as modern analogues of distributed molecular organization. (A) Contemporary hydrothermal vent ecosystems contain diverse archaeal, bacterial, eukaryotic, and viral communities that persist through metabolic cooperation, resource sharing, horizontal gene transfer, and adaptation to steep geochemical gradients. These environments provide continuous energy fluxes, mineral catalysts, and compartmentalized microenvironments that support complex ecological interactions. (B) Several network behaviors observed in hydrothermal vent communities—including energy capture, mineral-assisted catalysis, compartmentalization, information storage and exchange, distributed metabolism, and ecological adaptation—may be relevant to understanding transitions between prebiotic chemistry and cellular life. (C) Hypothetical Precellular Polymer Consortia (PPCs) are proposed as compartmentalized molecular ecosystems composed of interacting informational polymers, catalytic oligomers, metabolites, and environmental feedback systems. Although modern vent ecosystems are not direct descendants of PPCs, both systems illustrate how distributed interactions among many components can generate emergent properties, organizational continuity, and adaptive behavior. Hydrothermal vent communities therefore provide experimentally accessible models for investigating ecological and organizational principles that may have contributed to early evolution before LUCA.

Table 5. Extant microbial and molecular systems relevant to the SMN/PPC framework. These systems are not proposed as direct descendants of pre-cellular molecular ecosystems. Rather, they preserve organizational principles—including catalytic persistence, molecular memory, ecological cooperation, and distributed information processing—that may provide insight into evolutionary transitions preceding LUCA. ANME are anaerobic methane-reducing bacteria and SRB are sulfate-reducing bacteria.

Extant System	Key Feature	Relevance
Ribosome	Ancient catalytic RNA	Persistence of ancestral molecular architecture
RNase P	Catalytic ribozyme	Survival of RNA-based catalysis
CRISPR-Cas	Adaptive molecular memory	Information storage and retrieval
Hydrothermal vent archaea	Chemolithotrophic metabolism	Protometabolic organization
Syntrophic consortia (including ANME/SRB)	Distributed metabolism	Community-level organization
Microbial mats	Ecological specialization	Distributed adaptive networks

6.4. PLURIBUS and the Discovery of Unknown Hereditary Systems

The concepts developed throughout this review raise an important practical question: how can alternative hereditary systems be detected if they differ substantially from the DNA-, RNA-, and protein-based frameworks that dominate modern biology? Most contemporary molecular methods are optimized for well-characterized organisms and canonical biomolecules. As a result, our understanding of molecular diversity may be biased toward organisms that are readily cultured and experimentally tractable. If early evolution explored a broader range of hereditary chemistries than those preserved in modern biology, additional forms of informational molecules may remain undiscovered.

To address this challenge, we propose PLURIBUS (Planetary Liquid Universal Polymer Identification By Ultrasensitive Sequencing) as a conceptual framework for detecting and characterizing noncanonical informational polymers, molecular modifications, and hereditary networks. In the present framework, PLURIBUS is envisioned not primarily as a life-detection instrument for other worlds, but as a discovery platform for uncovering previously unrecognized hereditary chemistries within Earth’s microbial biosphere.

6.4.1. Extreme Environments as Natural Laboratories

Earth contains numerous environments that differ substantially from those occupied by traditional model organisms. Hydrothermal vent ecosystems, deep subsurface habitats, acidic hot springs, hypersaline lakes, polar environments, and other extreme ecosystems host diverse archaeal and bacterial communities that remain incompletely characterized. Many organisms inhabiting these environments are difficult to culture and consequently remain underrepresented in current molecular databases.

These ecosystems provide natural laboratories for investigating the limits of biological organization and molecular diversity. Hydrothermal vent archaea, chemolithotrophic microorganisms, and syntrophic consortia operate under conditions that may resemble aspects of early planetary environments. Such systems therefore represent attractive targets for discovering novel informational molecules, unusual catalytic mechanisms, and previously unrecognized forms of molecular memory. By focusing first on extreme terrestrial environments, PLURIBUS could establish a foundation for understanding the full range of hereditary chemistries that exist within the contemporary biosphere.

6.4.2. Beyond MODOMICS: Exploring Hidden Chemical Diversity

Current knowledge of RNA chemistry may represent only a fraction of the diversity present in nature. The MODOMICS database catalogs more than 300 naturally occur-

ring RNA modifications identified across a wide range of organisms [24,25]. However, many of these modifications were discovered through mass spectrometry and biochemical characterization of abundant RNAs obtained from readily cultured organisms [24,25]. Such approaches typically require substantial quantities of biological material and may therefore underrepresent rare modifications, environmentally induced modifications, and modifications present within uncultured microbial lineages.

This limitation may be particularly significant for archaea and extremophiles. Many archaeal groups inhabit environments that are difficult to sample and culture, while numerous microbial lineages remain known only through metagenomic surveys. Consequently, substantial regions of biological chemical space may remain unexplored.

Nanopore-based approaches offer an alternative strategy because they can potentially detect chemical modifications directly at the single-molecule level without requiring prior knowledge of their identities. Application of PLURIBUS-like approaches to archaeal and microbial ecosystems could therefore reveal modification chemistries that extend far beyond those currently represented in MODOMICS [24,25]. Such discoveries would expand our understanding of how molecular information can be stored, processed, and transmitted under diverse environmental conditions and may illuminate hereditary mechanisms that differ substantially from those observed in conventional model organisms.

6.4.3. From Earth Discovery to Astrobiology

Characterizing the full diversity of hereditary systems on Earth is a necessary prerequisite for searching for life elsewhere. Astrobiological investigations are inherently limited by assumptions derived from contemporary terrestrial biology. If alternative informational polymers, novel RNA modifications, or previously unrecognized hereditary systems exist within Earth's microbial biosphere, then current life-detection strategies may underestimate the diversity of biological possibilities.

PLURIBUS therefore provides a conceptual bridge between microbiology, origins-of-life research, and astrobiology. By first exploring extreme terrestrial ecosystems, the framework seeks to establish empirical benchmarks for recognizing unfamiliar hereditary systems. These insights could subsequently guide future investigations of potentially habitable environments beyond Earth, including Mars, Europa, Enceladus, Titan, and other planetary bodies.

More broadly, PLURIBUS extends the central themes of this review. If heredity can emerge from Stable Molecular Networks and Precellular Polymer Consortia rather than from canonical genomes alone, then future life-detection strategies should focus not only on specific biomolecules but also on the organizational properties of molecular systems. In this context, extant archaeal and microbial ecosystems become valuable testing grounds for understanding how alternative hereditary networks may arise, persist, and evolve across a wide range of planetary environments (Figure 6).

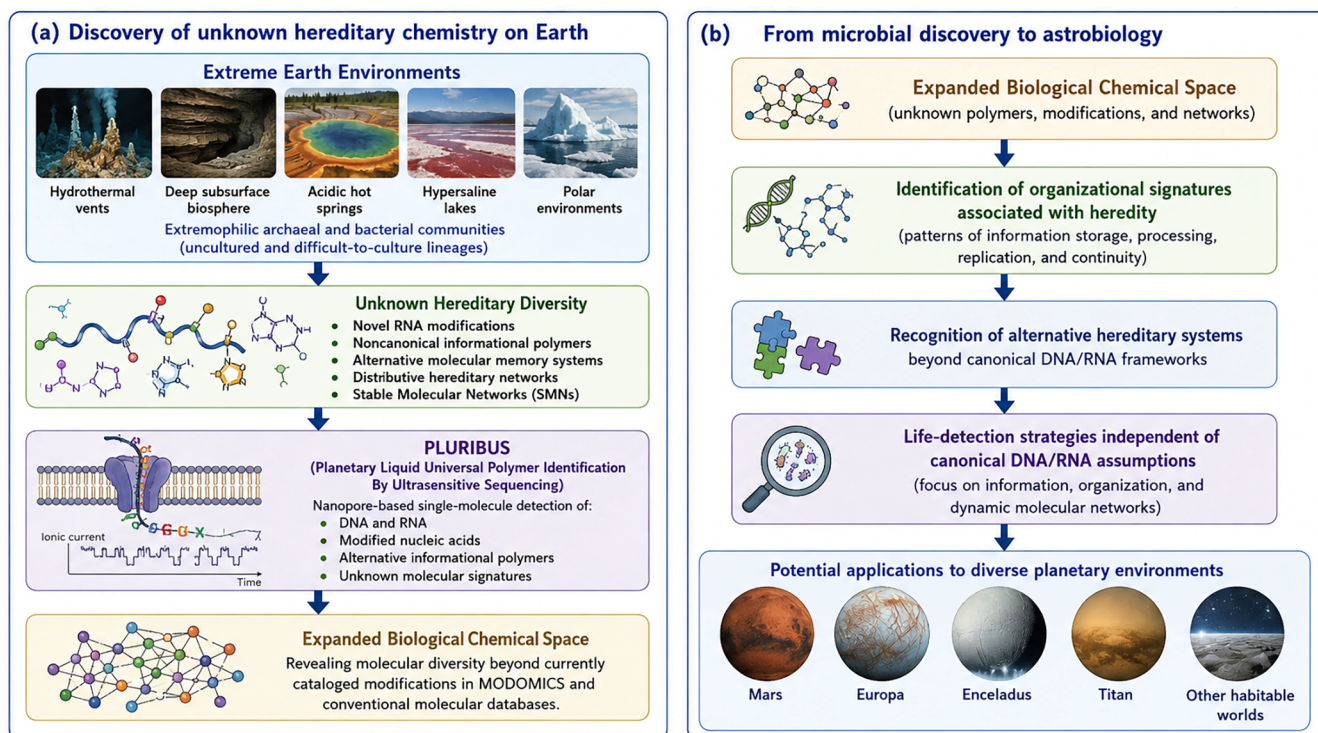


Figure 6. PLURIBUS as a framework for discovering alternative hereditary systems in extant microbial ecosystems and beyond. **(a)** Extreme terrestrial environments, including hydrothermal vents, deep subsurface habitats, acidic hot springs, hypersaline lakes, polar ecosystems, and other archaeal- and bacterial-dominated communities, may contain previously unrecognized forms of hereditary chemistry. These environments provide opportunities to identify novel RNA modifications, non-canonical informational polymers, alternative molecular memory systems, distributed hereditary networks, and Stable Molecular Networks (SMNs). PLURIBUS (Planetary Liquid Universal Polymer Identification By Ultrasensitive Sequencing) is proposed as a nanopore-based single-molecule discovery platform capable of detecting canonical and noncanonical informational molecules without requiring prior assumptions regarding their chemical composition. Such analyses may reveal biological chemical diversity extending beyond modifications currently cataloged in MODOMICS and conventional molecular databases. **(b)** Knowledge gained from extreme microbial ecosystems can be used to identify organizational signatures associated with heredity, including information storage, processing, replication, persistence, and network continuity. These discoveries may facilitate recognition of alternative hereditary systems that do not conform to conventional DNA/RNA-centered frameworks and support the development of generalized life-detection strategies based on organizational properties rather than specific biomolecules. Insights obtained from Earth's microbial biosphere may ultimately guide exploration of potentially habitable environments beyond Earth, including Mars, Europa, Enceladus, Titan, and other planetary bodies. The central concept of the PLURIBUS framework is that extant microbial ecosystems serve as experimental testing grounds for discovering previously unknown hereditary chemistries and molecular information systems, thereby providing a bridge between microbiology, origins-of-life research, and astrobiology.

7. Conclusions and Future Directions

The origin of life remains one of the most challenging problems in biology. Although the RNA World hypothesis has provided a valuable framework for understanding early evolution, increasing evidence suggests that heredity may have emerged through a broader range of molecular and organizational mechanisms than those preserved in contemporary biology. Alternative hereditary polymers, compositional inheritance, autocatalytic networks, and compartmentalized molecular systems collectively indicate that information

storage, amplification, and persistence are not necessarily restricted to modern DNA- and RNA-based genomes.

In this review, we proposed Stable Molecular Networks (SMNs) and Precellular Polymer Consortia (PPCs) as heuristic frameworks for exploring how organizational continuity might arise before the emergence of modern cells. These concepts build upon earlier theories of compositional inheritance, autocatalytic sets, communal evolution, and major evolutionary transitions by emphasizing interactions among informational polymers, catalytic systems, compartments, and environmental feedback processes. We further proposed that ecological diversification among PPCs could generate lineage-like evolutionary processes termed precellular speciation.

A central theme of this revised framework is that extant microbial systems may provide experimentally accessible models for investigating principles relevant to early evolution. CRISPR-Cas adaptive memory systems, ribosomes [29–31], RNase P [32–34], hydrothermal communities [39–41], thermophilic archaea, chemolithotrophic bacteria, microbial mats, and syntrophic consortia all demonstrate forms of molecular memory, distributed information processing, ecological cooperation, and organizational persistence. While none of these systems are proposed as direct descendants of pre-cellular molecular ecosystems, they illustrate biological principles that may have contributed to evolutionary transitions preceding LUCA.

Several predictions arising from the SMN/PPC framework are experimentally testable. Laboratory studies can examine whether interacting molecular populations exhibit organizational continuity, whether compartmentalized molecular communities can diversify under ecological selection, and whether network-level properties emerge that are not attributable to individual molecular components. Comparative analyses of CRISPR systems [29–31], ribosomal accretion patterns [32–34], RNase P evolution [32–34], microbial consortia, and hydrothermal vent ecosystems may further illuminate mechanisms by which information, metabolism, and ecological organization become integrated [39–41].

Finally, the possibility that life may arise through multiple hereditary architectures has important implications for astrobiology. If information storage, amplification, compartmentalization, and organizational persistence represent more fundamental properties of living systems than any molecular chemistry, then life elsewhere may differ substantially from terrestrial biology. The PLURIBUS framework was proposed to address this possibility by focusing on organizational signatures of heredity rather than specific biomolecules. Future astrobiological investigations may therefore benefit from searching for evidence of molecular memory, amplification, information exchange, compartmentalization, and network persistence in addition to canonical biosignatures.

Whether the earliest evolutionary systems resembled SMNs, PPCs, or entirely different organizational architectures remains unknown. Nevertheless, integrating insights from origins-of-life research, molecular evolution, microbiology, and astrobiology may help bridge the gap between planetary chemistry and cellular life. If heredity emerged from persistent molecular networks before the appearance of genomes and cells, then extant microbial systems may preserve organizational principles that illuminate one of the deepest transitions in the history of life.

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Abbreviations

The following abbreviations are used in this manuscript:

OoL	Origin of Life
PLURIBUS	Planetary Liquid Universal Polymer Identification By Ultrasensitive Sequencing
PPCs	Precellular Polymer Consortia
PIWI	P-element Induced WImpy testes
MODOMICS	RNA MODification OMICS
LUCA	Last Universal Common Ancestor
GARD	Graded Autocatalysis Replication Domain—Model for OoL
PPK	Preprokaryote
PNA	Peptide nucleic acid
TNA	Threose nucleic acid
GNA	Glycol nucleic acid
XNA	Xeno nucleic acid
SMN	Stable molecular network
ANA	Arabino nucleic acid
FANA	2-fluoro arabino nucleic acid
IOM	Insoluble inorganic matter
MOM	macromolecular organic matter
RAF theory	Reflexively Autocatalytic and Food-generated theory
CRISPR	Clustered Regularly Interspaced Short Palindromic Repeats
ANME	Anaerobic methane-reducing bacteria
SRB	Sulfate-reducing bacteria

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