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Question 5: Does the RNA-World Still Retain its Appeal After 40 Years of Research?

Pierre-Alain Monnard

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Abstract Forty years after its formulation, the hypothesis of the RNA-World remains rather controversial even though studies of RNA catalysis in cellular processes (for example, in the ubiquitous ribosomal peptide-bond formation) have clearly lent increased plausibility to the idea that an RNA-World existed at some point in the evolution leading to the emergence of cellular life. Indeed, several issues remain that weaken the concept: the synthesis of the RNA monomers under prebiotic conditions, their subsequent, efficient polymerization to yield ribozymes that specifically catalyze their own replication. This communication summarizes existing studies of the RNA polymerization from monomers. In our opinion, the recent developments show that given time plausible answers to some of the issues facing the RNA-World hypothesis will be found.

Keywords RNA-world · RNA polymerization · Nano-environment supported polymerization · Non-enzymatic catalysis

The origin(s) of a self-replicating genetic code represent(s) one inescapable issue to be resolved before a really plausible scenario for the emergence of Life on the early Earth can be defined. We are faced with several questions that can be listed in two main categories pertaining to the nature and primordial functions of an early genetic material and to the synthesis of its monomers and polymers.

In the recent years, the RNA-World hypothesis (Gilbert 1986; see Fig. 1) has been considered by many as a plausible scenario for the emergence of Life in part because of the complex nature of RNA that can act as a repository of genetic information, a genetic regulator, and a catalyst, as demonstrated in the laboratory by molecular selections (SELEX; Wilson and Szostak 1999), and observed multiple examples in contemporary cells (Winkler et al. 2004).

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P.-A. Monnard (✉)
Earth and Environmental Sciences EES-6, Los Alamos National Laboratory, Los Alamos, NM, USA
e-mail: pmonnard@lanl.gov

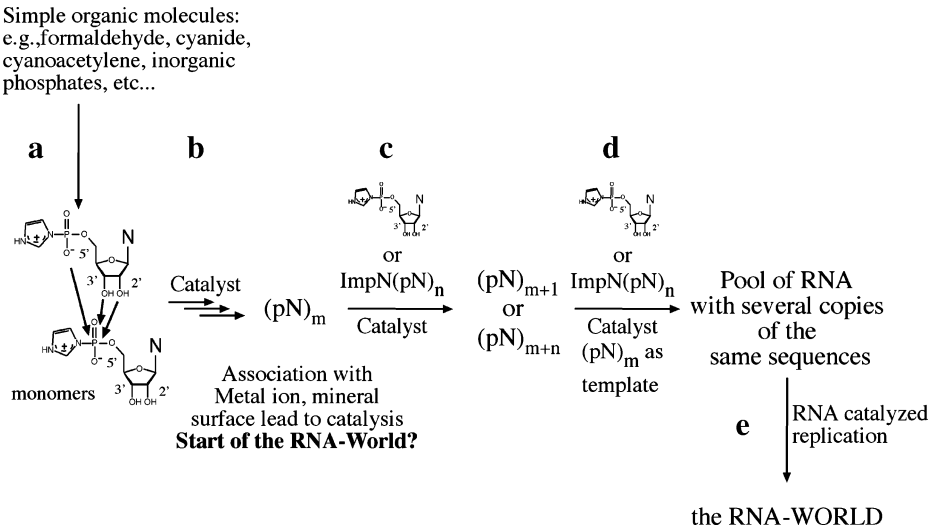

Fig. 1 Hypothetical route to the RNA-world, from the prebiotic RNA-monomer synthesis to the RNA-catalyzed, template-directed RNA replication. *N* stands for any nucleobase, *(pN)* for a RNA oligomer or polymer, *ImpN(pN)* for an activated RNA oligomer or polymer, *m*, *m*+1 and *m*+*n* subscripts refer to the lengths of a polymer. **a** Synthesis of the activated RNA monomers from simple organic precursors. In the laboratory, imidazole often serves as the activating group though it seems improbable on the early Earth. This phase relies on the prebiotic chemistry. **b** Oligomerization of the monomers. Monomers are incorporated into oligomers in the presence of a catalyst, such as mineral surface, dissolved metal ions or nano-structured media (eutectic phase in water ice or matrices formed by amphiphile bilayers). At that stage, RNA fragments associated with metal ions might already enhance their own duplication. One could consider such a process the beginning of the RNA world. **c** Synthesis of longer polymers. RNA activity is related to the sequence of its constituent monomers and the conformation the resulting polymer can assume. A minimal sequence length is therefore required to permit catalytic activity. Long polymers can be obtained either by ligation of the previously formed oligomers that were activated or by further monomer addition. **d** Non-enzymatic, template-directed polymerization. To transmit the sequence of catalytic RNA fragments, the ability to non-enzymatically copy these molecules is essential, even though it might not initially be as accurate as today's enzymatic machinery. This process will not only permit an increase of the catalytic activity, but also of the RNA-pool size, as well as the emergence of new catalysts because of copying errors. **e** The RNA World. At same point, the RNA pool will contain molecules that are also catalysts of their own copying, or replication, as well as those of any other functional RNAs. A full-fledged RNA world is then achieved

However, this view has been challenged because of issues intrinsic to RNA chemistry: (1) the prebiotic synthesis of its complex monomers which requires the stereospecific linkage of the nucleobase on a ribose sugar itself linked to a phosphate group (a difficult proposition that would seemingly result in an exceedingly low monomer concentration); (2) the regiospecificity of the phosphoester bonds; and (3) the relative fragility of long RNA polymers in aqueous solutions. Other objections raised against the RNA world hypothesis pertain to the activities of RNA catalysts, i.e. to the mechanisms that must have led to the emergence of specific, rather long, active sequences over all possible sequences. Indeed, if a RNA fragment length of 30 is assumed to be required for a catalytic activity, approximately 10^{18} different sequences (0.02 g RNA) are possible and could be synthesized (Monnard 2005; Orgel 2004); if a 50-mer length is needed, 10^{30} RNAs (3.5×10^{7} kg RNA) theoretically exist, but cannot likely be made at once. Although several sequences could have had a similar activity, concentration issues associated with the total catalytic activity must not be overlooked. Can the RNA World retain its appeal?

We only partially agree with the caveats presented above because some of the most recent studies seem to indicate directions for the resolution of these issues. Benner's group has shown that ways exist to concentrate ribose on borate from all sugars produced in the formose reaction (Ricardo et al. 2004) and the synthesis of the four nucleobases and more isomers under prebiotic conditions has been achieved (Ferris 2005). However, the complete synthesis of RNA monomers still remains elusive because ribose synthesis and phosphorylation reactions happen under very different conditions that seem today to be irreconcilable.

Polymerization of chemically activated natural nucleotide monophosphates on clays (Huang and Ferris 2006) and in the eutectic phase in water–ice (Monnard et al. 2003) and more recently in lipid-bilayer lattices (Rajamani et al. 2007) under plausible prebiotic conditions has yielded relatively long oligomers 20 to 50-mers in significant amounts. Moreover, these methods seem to at least partially resolve the issue of low starting monomer concentrations: indeed, in all three cases, the monomers are concentrated from dilute solutions and are ordered in an assembly conducive to efficient polymerization. On clays (Miyakawa and Ferris 2003) and in our work on the eutectic phase in water–ice, it was established that the elongation of oligomers by monomer addition can be dependent on the oligomer sequences: e.g., in the eutectic phase in water–ice, long single stranded RNA fragments are only efficiently elongated by monomer addition up to 30-mer in length whereas sequences capable of forming short double stranded structures such as hairpins can be extended up to 40 to 45-mers in length (Monnard and Szostak, unpublished). This indicates that all sequences are not equally probable.

Several of our preliminary results also show that non-enzymatic, template-directed replication of a nucleic acid strand should proceed by Watson–Crick base pairing with every nucleobase, even uridine, being incorporated at similar rates in the eutectic phase in water–ice (Monnard and Szostak, unpublished).

Of course, the RNA-World still requires an elusive RNA-polymerase ribozyme (thought here in the broad sense of a ligase) with the capacity of replicating itself at rates that surpass its decomposition rate. Assuming a pair of RNA polymerases existed (one template and one replicator), we believe that they alone would have not permitted the emergence of RNA "life." Indeed, to consider the emergence and evolution of a genetic RNA code alone seems out of context (Szostak et al. 2001). Life, as we know it, suggests that, very early on, membranous and metabolic structures would have been necessary to create a protocell that only survived because of their synchronous action and reciprocal feed-back regulation. A compartment would have played an essential role by permitting the accumulation and maintenance of high concentrations of reacting species, as well as the distinct evolution of a particular protocell.

Whether or not one believes in the RNA World and further considers RNA as the actual first genetic material, the RNA model in the current absence of any other plausible precursor represents a system that allows us to explore essential aspects of the emergence of a polymeric, genetic system without the requirement of a complex metabolism.

References

Ferris JP (2005) Catalysis and prebiotic synthesis. Rev Mineral Geochem 59:187–210

Gilbert W (1986) Origin of life: the RNA world. Nature 319:618

Huang W, Ferris JP (2006) One-step, regioselective synthesis of up to 50-mer of RNA oligomers by montmorillonite catalysis. J Am Chem Soc 128:8914–8919

Miyakawa S, Ferris JP (2003) Sequence- and regioselectivity in the montmorillonite-catalyzed synthesis of RNA. J Am Chem Soc 125:8202–8208

Monnard P-A (2005) Catalysis in abiotic structured media: an approach to selective synthesis of biopolymers. Cell Mol Life Sci 62(5):520–534

Monnard P-A, Kanavarioti A, Deamer DW (2003) Eutectic phase polymerization of activated ribonucleotide mixtures yields quasi-equimolar incorporation of purine and pyrimidine nucleobases. J Am Chem Soc 125:13734–13740

Orgel LE (2004) Prebiotic chemistry and the origin of the RNA world. Crit Rev Biochem Mol Biol 39(2):99–123

Rajamani S, Vlassov A, Coombs A, F. O, Deamer DW (2007) Lipid-assisted synthesis of RNA-like polymers from mononucleotides. Orig Life Evol Biosphere (in press)

Ricardo A, Carrigan MA, Olcott AN, Benner SA (2004) Borate minerals stabilize ribose. Science 303:196

Szostak JW, Bartel DP, Luisi PL (2001) Synthesizing life. Nature 409:387–390

Wilson DS, Szostak JW (1999) In vitro selection of functional nucleic acids. Ann Rev Biochem 68:611–647

Winkler WC, Nahvi A, Collins JA, Breaker RR (2004) Control of gene expression by a natural metabolite-responsive ribozyme. Nature 428:281–286