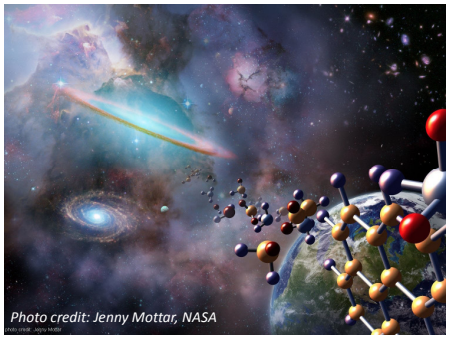
The molecular origins of life



WS 2016

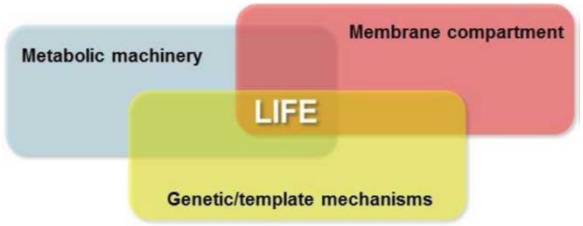
Zbigniew Pianowski

Origin of the Universe – stars, planets, elements

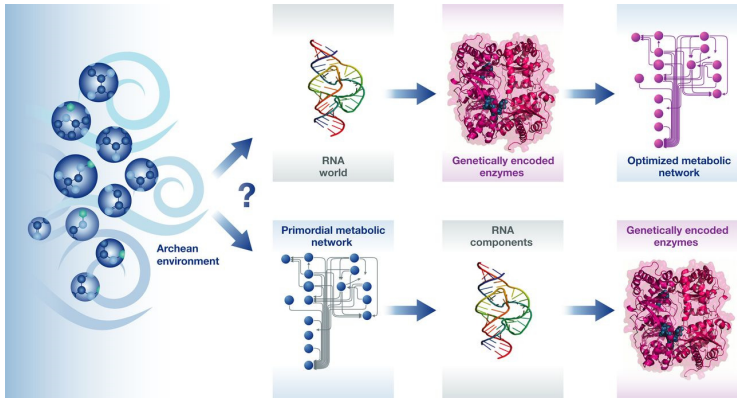
Origin of biorelevant monomers – primordial soup

Complex chemical processes on the way to living systems

Protocells and LUCA



Route to life by chemical networks



P. L. Luisi *Mol Syst Biol.* **2014,** *10,* 729

Self-organization of chemical networks

One of pre-conditions for life is to be far from thermodynamic equilibrium.

Life uses non-linear effects to amplify and stabilize minor environmental effects

Spatial and temporal synchronisation of reactive processes provides molecules with patterns of collective behavior
Under certain conditions far from thermodynamic equilibrium, heterogenous mixtures can trigger emergent properties at the collective level.

Oscilatory and autocatalytic processes are very common in biological systems.
Examples include: metabolic cycles, immune response, or apoptosis.

Oscilatory reactions – importance for homeostasis. Provide positive and negative feedback loops to maintain the dynamic far-from-equilibrium state of the system.

Self-organization and self-assembly processes are under tight enzymatic control in all living organisms.
However, oscilatory and autocatalytic behavior can appear sponateously in much simpler molecular systems.

Oscilatory reactions in biology

Endogenous processes - arise from feedbacks and internal loops between the different components of metabolic networks
ATP/ADP concentration in glycolytic cycle, circadian oscilations, metabolic rhytms, sleep-wake cycle

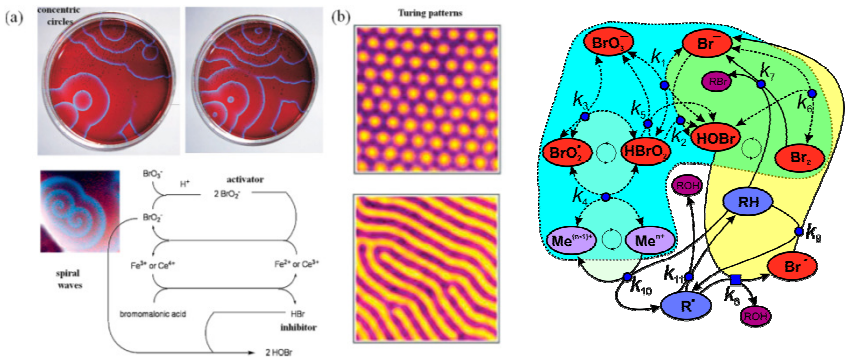
Exogenous processes – arise from external fluctuations in the environment
temperature, pH, humidity, illumination, UV irradiation, astronomic cycles

Chemical systems that mimic biological oscilations are studied as simple models

Belousov-Zhabotynski, CIMA reaction

Oscilatory reactions – activation and inhibition steps provide feedback loops to control the reaction speed.
The most ancient protometabolic networks could have similar basic properties.

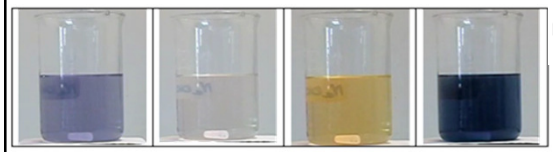
Belousov-Zhabotynski (BZ) reaction



The reaction usually involves potassium bromate(VII) and malonic acid, optionally with cerium(IV) sulfate and citric acid. Ferroin is one of the common redox indicator

Briggs-Rauscher reaction

$IO_3^- + 2\,H_2O_2 + CH_2(CO_2H)_2 + H^+ \rightarrow ICH(CO_2H)_2 + 2\,O_2 + 3\,H_2O$

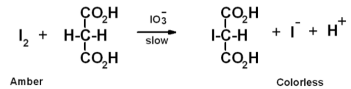


1. **The iodate ion is changed into iodine by hydrogen peroxide. The color changes to amber:**

$$2\,IO_3^- + 2\,H^+ + 5\,H_2O_2 \xrightarrow[\text{fast}]{Mn^+} I_2 + 5\,O_2 + 6\,H_2O$$

Colorless → Amber

2. **The free iodine reacts with malonic acid to produce iodide ions.**



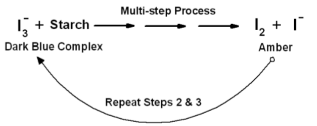
Amber → Colorless

3. **The free iodine combines with iodide very rapidly to form the negative ion I_3^-, which reacts with starch to form a dark blue complex:**

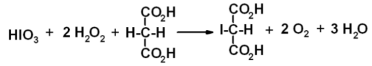
$$I_2 + I^- \xrightarrow[\text{fast}]{} I_3^- + \text{Starch} \xrightarrow[\text{fast}]{} \text{Dark Blue Complex}$$

Amber

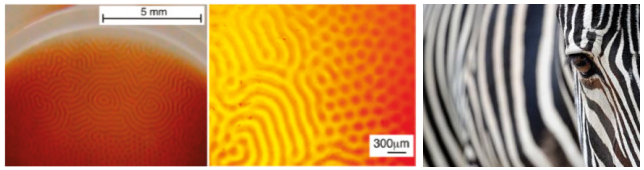
4. **After a period of time, the I_3^- ions are converted back into iodine and iodide ions, so the dark blue color disappears and the process repeats itself:**



5. **Eventually the faster step 3 becomes dominant and the change of I_3^- back to iodine/iodide stops after about 15 cycles, so the solution remains dark blue. The overall chemical reaction is:**



Chlorite/iodide/malonic acid (CIMA) reaction



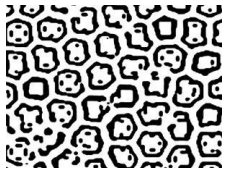
For the spontaneous generation of a Turing pattern, two intermediate species, an activator and an inhibitor, should be generated with the diffusion coefficient of the activator smaller than that of the inhibitor. The CIMA reaction that generates the activator, I^-, and inhibitor, ClO_2^-, was performed in an open gel reactor.

The mechanism of Turing pattern generation is also likely responsible for formation of stripes in certain mammals (e.g. zebra), or arrangement of leafs in plants

J. Phys. Chem. B 115(14):3959-63

Turing patterns also observed in metabolic reactions (glycolysis)

PLoS ONE 2007, 2(10):e1053



„Rosette" spots of a jaguar can be reproduced by two coupled activator/inhibitor processes

Autocatalytic processes

Inherent components of oscilatory reactions

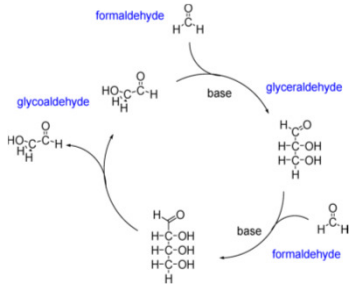
Explain the origin of homochirality

Fundamental concept for any system that grows and produces more copies of itself

Transition from chemical systems to biological ones inherently involves autocatalysis

Particularly interesting are links between chemistry and primitive metabolic pathways

Autocatalytic processes – formose reaction

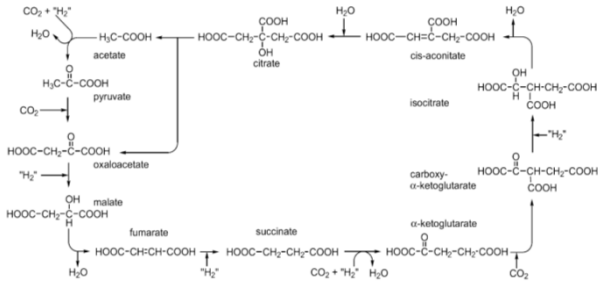


Formose reaction is one of the simplest autocatalytic cycles – two molecules of glycolaldehyde are produced from one.

Such unitary autocatalytic cycles would provide kinetic evolutionary advantage to evolving metabolic networks

Prebiotic variants of the reductive citric acid (Krebs/tricarboxylic acid) cycle

TCA/Krebs cycle is central for metabolism in aerobic forms of life.
The reverse citric acid cycle is used by some bacteria to produce complex carbon compounds from CO_2 and H_2O

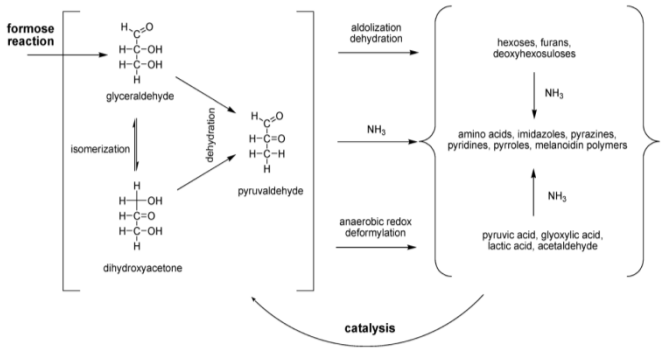


This catalytic cycle is claimed (Morowitz) to be able to run also in absence of enzymes (e.g. on mineral surfaces).
This could be the starting point for evolution of all other currently operating metabolic cycles.
However, no experimental demonstration of the full cycle under abiotic conditions delivered yet.

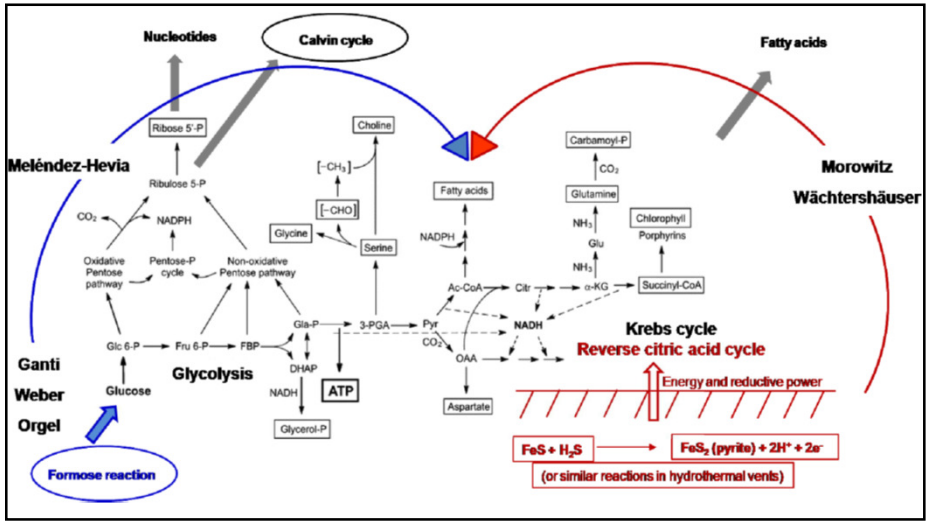
Problems: cross-reactivity, side reactions that drain active intermediates and energy until cycles stop.

More complex views on autocatalytic cycles

Coupling formose reaction with ammonia and thiols yields reactive α-hydroxy and α-aminothioesters, as well as numerous other aliphatic and aromatic compounds. Some of them enter another autocatalytic cycles.

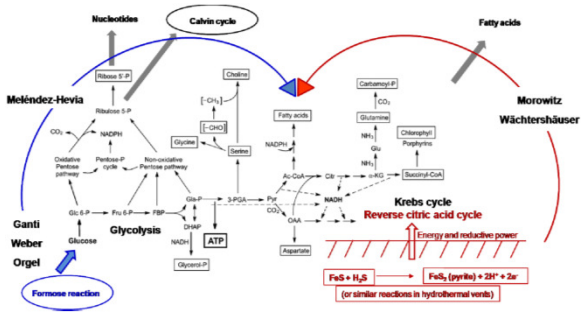


This additionally suggests that glycolysis was the ancient metabolic pathway

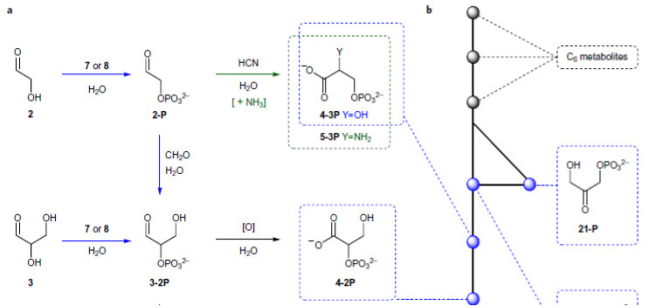


More complex views on autocatalytic cycles

(Black) Minimal metabolic map, constructed by simplifying present-day cellular metabolisms. (Blue) The clockwise sense of metabolic evolution in the scheme of Meléndez-Hevia et al.497 gives the formose reaction a prominent role as the first metabolic cycle, as Weber, Meléndez- Hevia, or Ganti proposed. (Red) The counterclockwise sense of metabolic evolution, according to the same scheme, would come from considering the reverse citric acid cycle as the first metabolic cycle, as Morowitz or Wächtershäuser have defended. In that case, energy and reductive power could be provided by redox reactions occurring on mineral surfaces (e.g., FeS, NiS) in hydrothermal vents, for instance.

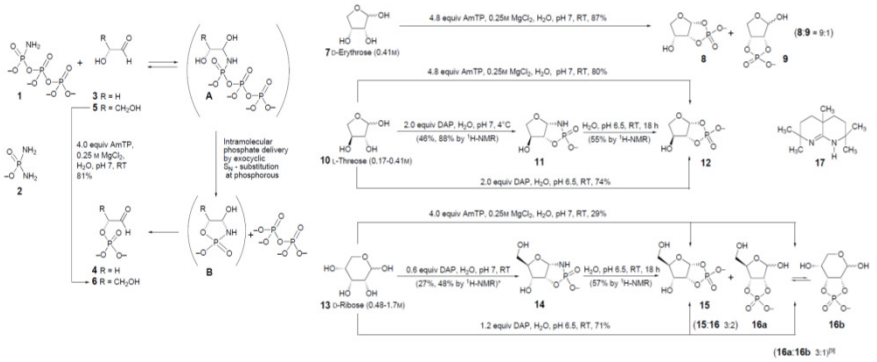


Phosphoenolpyrruvate – important metabolic intermediate



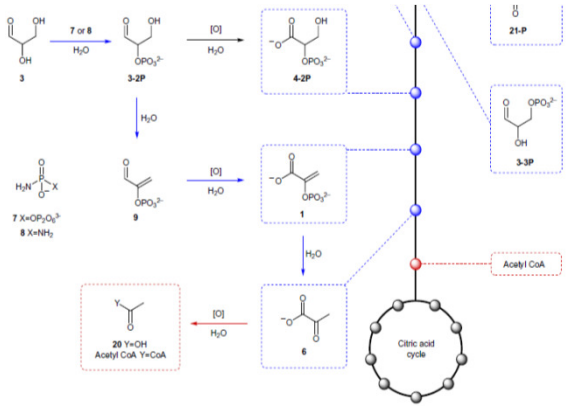
A. Coggins, M. Powner *Nature Chem.* **2016**, *DOI: 10.1038/NCHEM.2624*

Phosphorylation of sugars



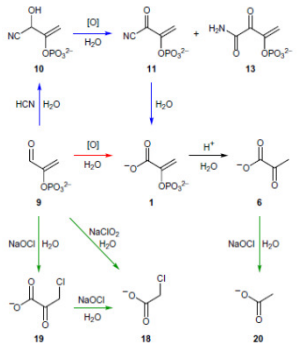
A. Eschenmoser, *et al. Angew. Chem. Int. Ed.* **2000**, *39*, 2281-2285

Phosphoenolpyrruvate – important metabolic intermediate



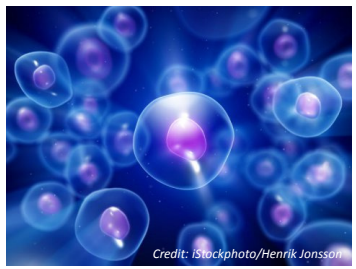
A. Coggins, M. Powner *Nature Chem.* **2016**, *DOI: 10.1038/NCHEM.2624*

Phosphoenolpyrruvate – important metabolic intermediate



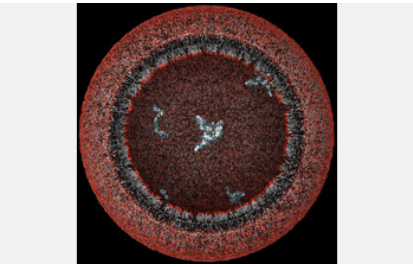
A. Coggins, M. Powner *Nature Chem.* **2016**, *DOI: 10.1038/NCHEM.2624*

Encapsulation – essential for life



Membrane compartments

Assembly of amphiphilic monomers into protocellular compartments

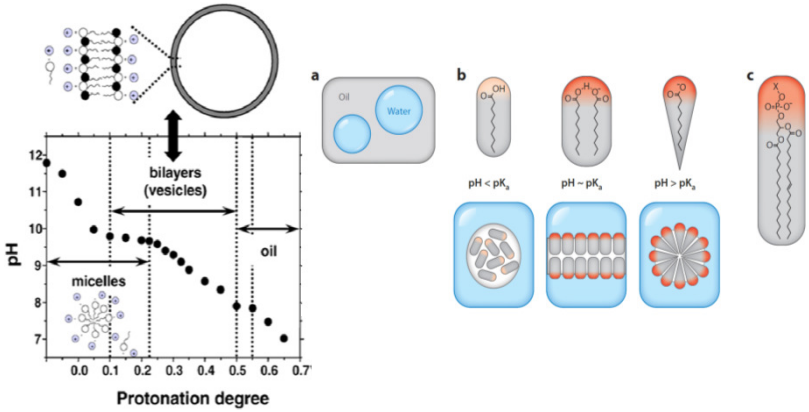


Credit: *Janet Iwasa*

A three-dimensional view of a model protocell (a primitive cell) approximately 100 nanometers in diameter.

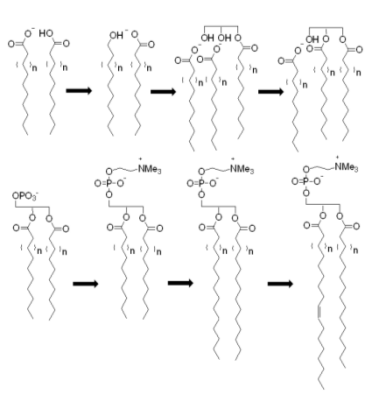
The protocell's fatty acid membrane allows nutrients and DNA building blocks to enter the cell and participate in non-enzymatic copying of the cell's DNA. The newly formed strands of DNA remain in the protocell

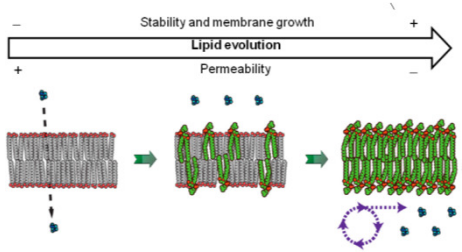
pH-dependent phase behavior of fatty acids in water



80 mM oleic acid/ sodium oleate in water

Scheme of the membrane evolution

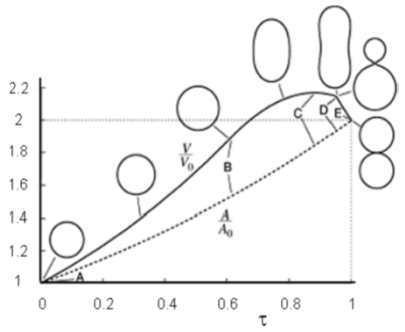


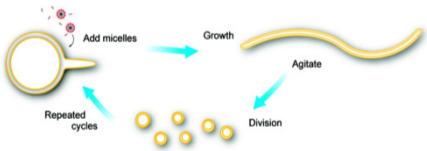


More complex components lead to slower amphiphile desorption and thus faster growth of the protocell. Decreasing permeability is a selective pressure for the emergence of internalized metabolic and transport machinery in the system

Chemical evolution of membrane components

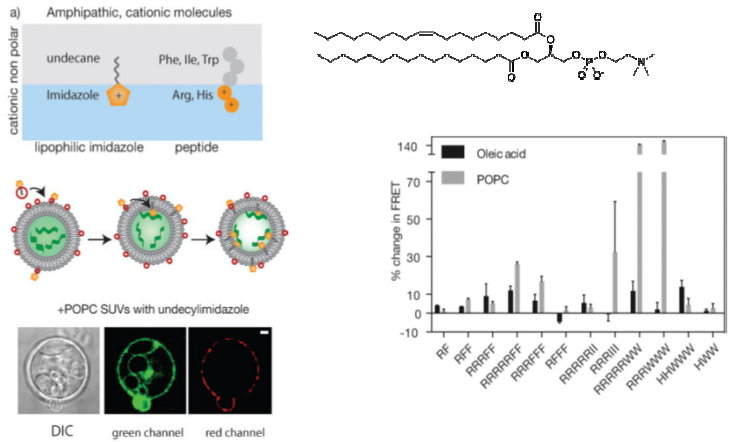
Growth and division of vesicles





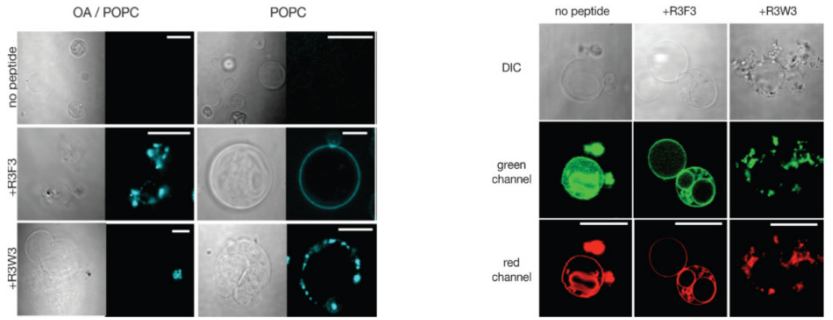
Ting F. Zhu, and Jack W. Szostak *J. Am. Chem. Soc.*, **2009**, 131 (15), 5705-5713

Noncovalent nucleotide association with membranes



Neha P. Kamat, Sylvia Tobe, Ian T. Hill, and Jack W. Szostak *Angew. Chem. Int. Ed.* **2015**, *54*, 11735 –11739

Noncovalent nucleotide association with membranes

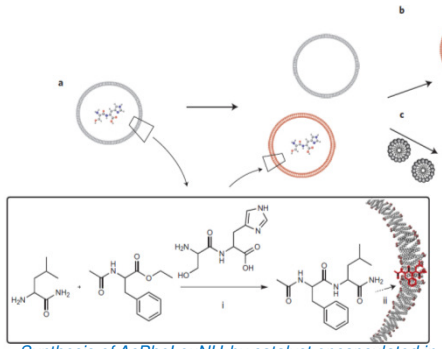


Neha P. Kamat, Sylvia Tobe, Ian T. Hill, and Jack W. Szostak *Angew. Chem. Int. Ed.* **2015**, *54*, 11735 –11739

Adaptive changes and competition between protocell vesicles

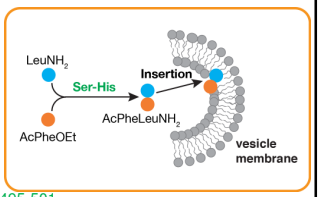
Vesicles with $AcPheLeuNH_2$ in the membrane (red) grow when mixed with vesicles without dipeptide (grey), which shrink

After micelle addition vesicles with $AcPheLeuNH_2$ in the membrane grow more than vesicles without the dipeptide.



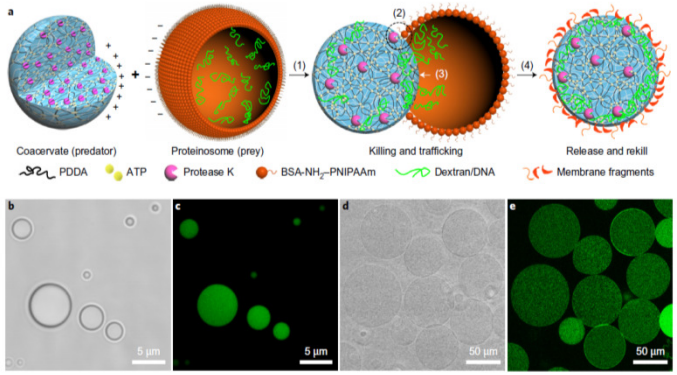
Synthesis of $AcPheLeuNH_2$ by catalyst encapsulated in fatty-acid vesicles. The dipeptide Ser-His catalyses the reaction between substrates $LeuNH_2$ and AcPheOEt (i), which generates the product of the reaction, $AcPheLeuNH_2$.

The product dipeptide $AcPheLeuNH_2$ localizes to the bilayer membrane



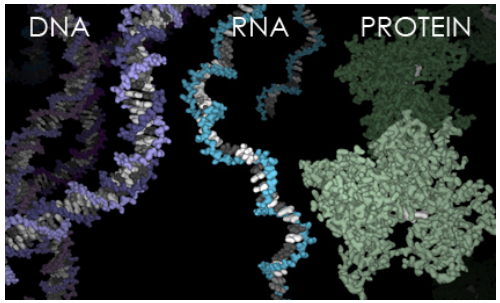
K. Adamala, J. W. Szostak *Nature Chem.* **2013**, *5*, 495-501

Predator/prey behavior



S. Mann *et al. Nature Chem.* **2016**, *DOI: 10.1038/NCHEM.2617*

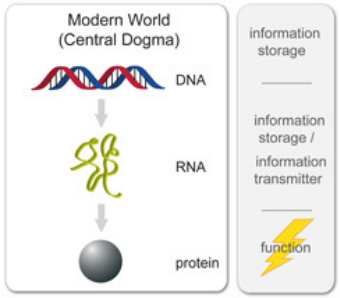
„Genes-first"



In modern cells, RNA (light blue, center) is made from a DNA template (purple, left) to create proteins (green, right).

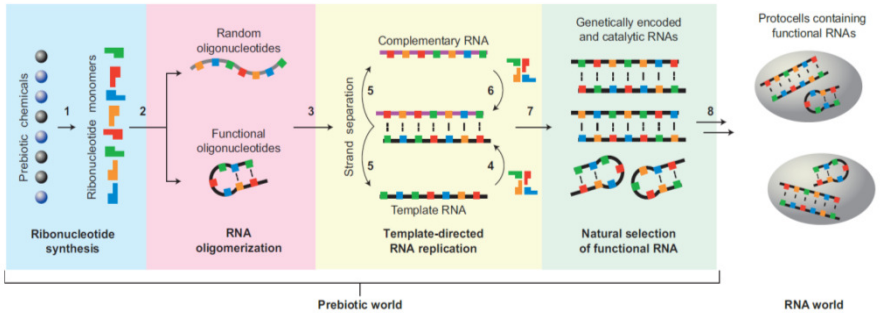
RNA folding is mediated by base-pairing interactions along different regions of a single-stranded RNA.

The RNA world

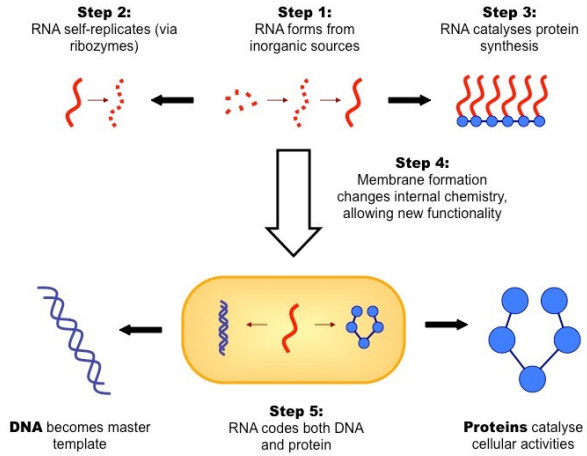


Conceptual idea that there was a period in the early history of life on Earth when RNA (or its structurally simplified analogue) carried out most of the information processing and metabolic transformations needed for biology to emerge from chemistry

The RNA world



The RNA world

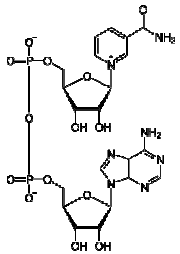


The RNA world

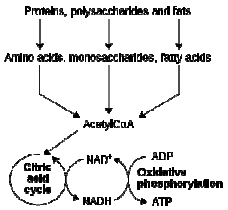
Crick, Orgel and Woese speculated in 1968 that, because RNA can form secondary structures, it has both a genotype and a phenotype and is a good candidate for the emergence of life

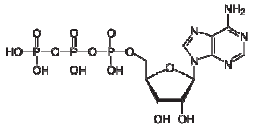
F. H. C. Crick *J. Mol. Biol.* **1968**, *38*, 367-379, L. E. Orgel *J. Mol. Biol.* **1968**, *38*, 381-393

Ribonucleotide coenzymes currently used by many proteins may be molecular „fossils" from the primoridal RNA-based metabolism



Nicotinamide adenine dinucleotide (NAD$^+$)



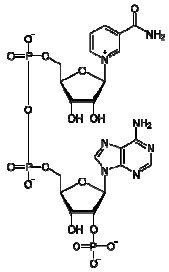


Adenosine triphosphate (ATP)

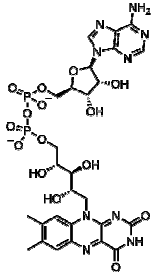
H. B. White III *J. Mol. Evol.* **1976**, *7*, 101-104

The RNA world

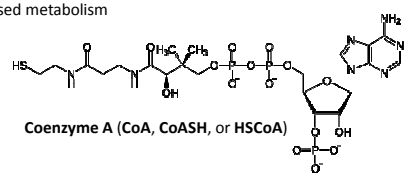
Ribonucleotide coenzymes now used by many proteins may be molecular „fossils" from the primoridal RNA-based metabolism



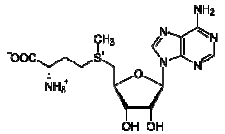
Nicotinamide adenine dinucleotide phosphate (NADP$^+$)



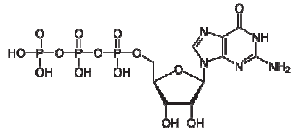
flavin adenine dinucleotide (FAD)



Coenzyme A (CoA, CoASH, or HSCoA)



***S*-Adenosyl methionine**

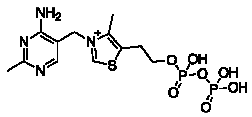


Guanosine-5'-triphosphate (GTP)

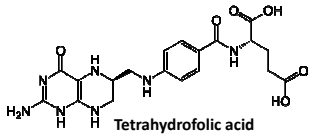
H. B. White III *J. Mol. Evol.* **1976**, *7*, 101-104

The RNA world

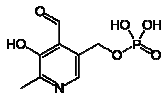
Other coenzymes contain cyclic nitrogen-containing bases that can also derive from nucleotides



Thiamine pyrophosphate (TPP or ThPP) – Vit. B_1



Tetrahydrofolic acid



Pyridoxal phosphate (PLP) – Vit. B_6

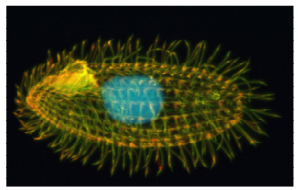
H. B. White III *J. Mol. Evol.* **1976**, *7*, 101-104

The RNA world

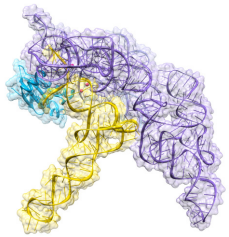
Ribozymes – Ribonucleic acid enzymes

1989 – Thomas Cech and Sidney Altman – Nobel Prize in chemistry for discovery of catalytic RNA

Thomas R. Cech was studying RNA splicing in the ciliated protozoan *Tetrahymena thermophila*
Sidney Altman and Norman Pace were studying the bacterial RNase P complex.



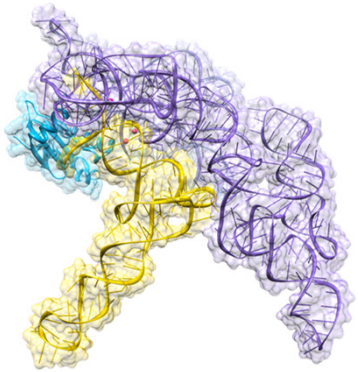
Tetrahymena thermophila



Bacterial RNAse P

The RNA world

Ribonuclease P

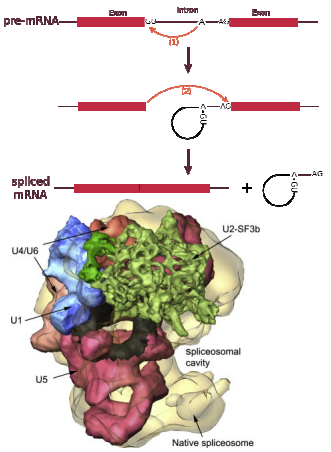


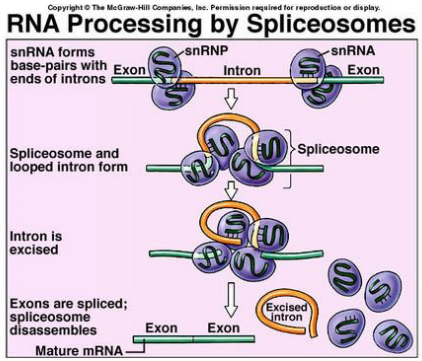
Ribonuclease P (RNase P) is a type of ribonuclease which cleaves RNA. RNase P is unique from other RNases in that it is a ribozyme – a ribonucleic acid that acts as a catalyst in the same way that a protein based enzyme would. Its function is to cleave off an extra, or precursor, sequence of RNA on tRNA molecules.

Bacterial RNase P has two components: an RNA chain, called M1 RNA, and a polypeptide chain, or protein, called C5 protein. *In vivo*, both components are necessary for the ribozyme to function properly, but *in vitro*, the M1 RNA can act alone as a catalyst. The primary role of the C5 protein is to enhance the substrate binding affinity and the catalytic rate of the M1 RNA enzyme probably by increasing the metal ion affinity in the active site.

Crystal structure of a bacterial ribonuclease P holoenzyme in complex with tRNA (yellow), showing metal ions involved in catalysis (pink)

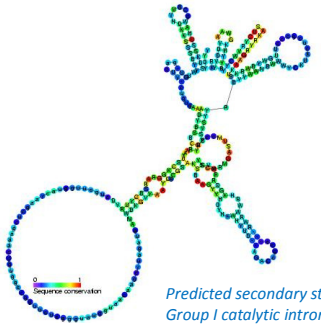
RNA splicing





Spliceosome – a complex of ribonucleoproteins

RNA splicing

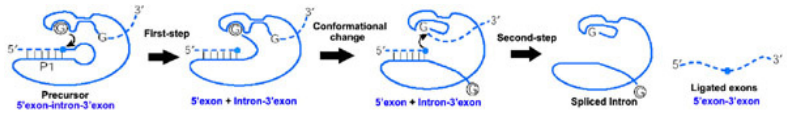


Self-splicing RNA introns

RNA splicing in *Tetrahymena* was taking place also in absence of the spliceosome - the ‚negative control' obtained after protease digestion also spliced.

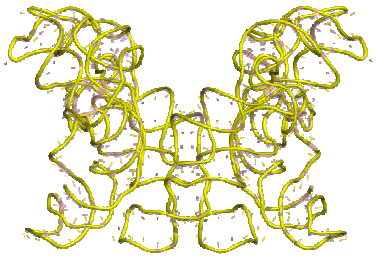
In contrary to the spliceosome, the ***catalytic*** motif ***does not*** contain protein part, ***only RNA***.
First known example of a ***ribozyme*** – ribonucleic acid-composed enzyme analogue.

Predicted secondary structure and sequence conservation of Group I catalytic intron



RNA splicing

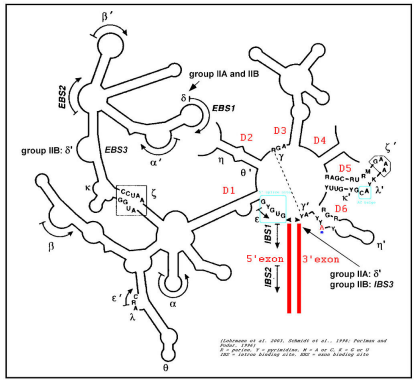
Group I catalytic introns



A 3D representation of the Group I catalytic intron.
This view shows the active site in the crystal structure of the Tetrahymena ribozyme

RNA splicing

Group II catalytic introns

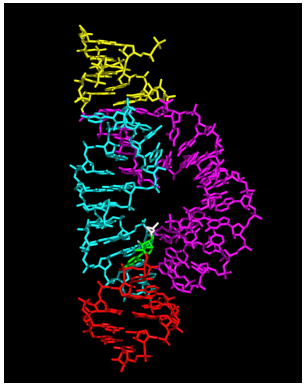


Ribozyme activity (e.g., self-splicing) can occur under high-salt conditions in vitro. However, assistance from proteins is required for in vivo splicing

It is hypothesized that pre-mRNA splicing may have evolved from group II introns, due to the similar catalytic mechanism as well as the structural similarity of the Domain V substructure to the U6/U2 extended snRNA

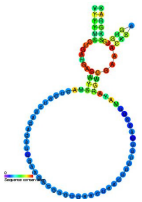
Ribozymes

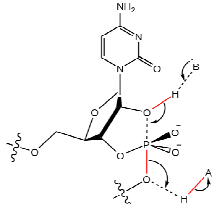
Hammerhead ribozyme



The hammerhead ribozyme is a RNA molecule motif that catalyzes reversible cleavage and joining reactions at a specific site within an RNA molecule.

- model system for research on the structure and properties of RNA,
- targeted RNA cleavage experiments,



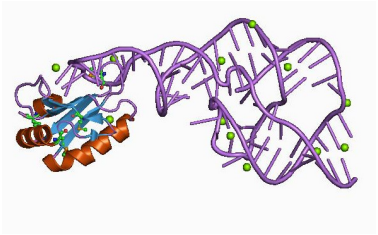


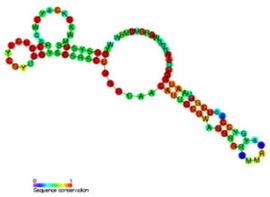
Ribozymes

HDV ribozyme

The hepatitis delta virus (HDV) ribozyme is a non-coding RNA found in the hepatitis delta virus that is necessary for viral replication and is thought to be the only catalytic RNA known to be required for viability of a human pathogen.

The ribozyme acts to process the RNA transcripts to unit lengths in a self-cleavage reaction. The ribozyme is found to be active in vivo in the absence of any protein factors and is the fastest known naturally occurring self-cleaving RNA.





Riboswitches

Riboswitches demonstrate that naturally occurring RNA can bind small molecules specifically.

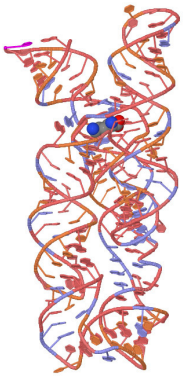
Before discovery of riboswitches only proteins were supposed to do so in the biological context.

Riboswitches exist in all domains of life, and therefore are likely that they might represent ancient regulatory systems or fragments of RNA-world ribozymes whose binding domains remained conserved throughout the evolution

A riboswitch is a regulatory segment of a messenger RNA molecule that binds a small molecule, resulting in a change in production of the proteins encoded by the mRNA.

The discovery that modern organisms use RNA to bind small molecules, and discriminate against closely related analogs, expanded the known natural capabilities of RNA beyond its ability to code for proteins, catalyze reactions, or to bind other RNA or protein macromolecules.

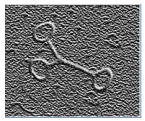
Most known riboswitches occur in bacteria, but functional riboswitches of one type (the TPP riboswitch) have been discovered in plants and certain fungi. TPP riboswitches have also been predicted in archaea, but have not been experimentally tested.



The lysine riboswitch

Viroids

Viroids ("subviral pathogens„) are mostly plant pathogens, which consist of short stretches of highly complementary, circular, single-stranded, and non-coding RNA without a protein coat.
Viroids are extremely small - 246 to 467 nucleobases (genomes of smallest viruses start from 2,000 nucleobases).
Viroids are plausible "living relics" of the RNA world.



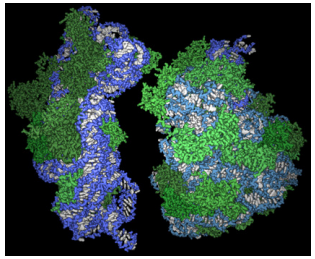
PSTVd-infected potatoes (right)

Pertinent viroid properties listed in 1989 are:
their small size, imposed by error-prone replication;
their high guanine and cytosine content, which increases stability and replication fidelity;
their circular structure, which assures complete replication without genomic tags;
existence of structural periodicity, which permits modular assembly into enlarged genomes;
their lack of protein-coding ability, consistent with a ribosome-free habitat; and
replication mediated in some by ribozymes—the fingerprint of the RNA world.

Putative secondary structure of the PSTVd viroid

Ribosome – the ‚smoking gun'

Ribosome is a ribozyme!



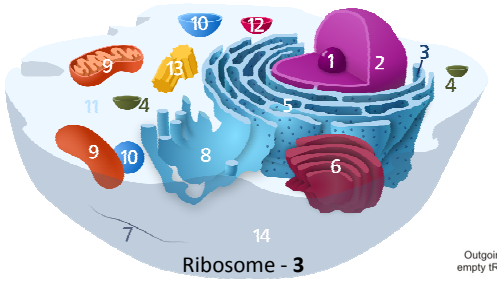
Ribosome: green - proteins, blue and white - RNA

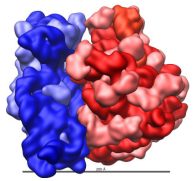
The ribosome is a simple molecular machine, found within all living cells, that serves as the site of biological protein synthesis (translation). Ribosomes link amino acids together in the order specified by messenger RNA (mRNA) molecules.

Ribosome is structurally highly conserved among all living species – most likely present in LUCA

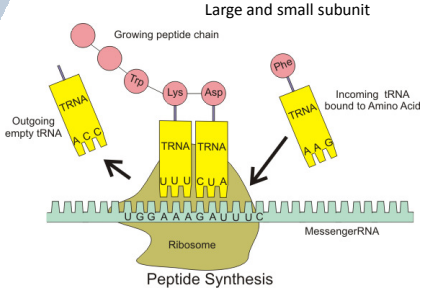
Ribosomes consist of two major components: the small ribosomal subunit, which reads the RNA, and the large subunit, which joins amino acids to form a polypeptide chain. Each subunit is composed of one or more ribosomal RNA (rRNA) molecules and a variety of ribosomal proteins.

Ribosome – the ‚smoking gun'





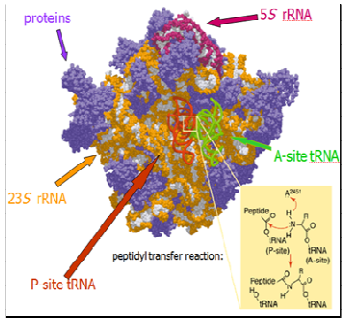
Large and small subunit



Peptide Synthesis

Ribosome – the ‚smoking gun'

Ribosome is a ribozyme!



No protein is present within 18 Angstroms from the active site → proteins play a structural role, but
DO NOT CATALYZE THE ACYL TRANSFER PROCESS

T. Cech *Science*. **2000**, *289*, 878-879

Ribosome – the ‚smoking gun'

Ribosome is a ribozyme!

The ribosome may have first originated in an RNA world appearing as a self-replicating complex that only later evolved the ability to synthesize proteins when amino acids began to appear.

Studies suggest that ancient ribosomes constructed solely of rRNA could have developed the ability to synthesize peptide bonds.

In addition, evidence strongly points to ancient ribosomes as self-replicating complexes, where the rRNA in the ribosomes had informational, structural, and catalytic purposes because it could have coded for tRNAs and proteins needed for ribosomal self-replication.

As amino acids gradually appeared in the RNA world under prebiotic conditions, their interactions with catalytic RNA would increase both the range and efficiency of function of catalytic RNA molecules. Thus, the driving force for the evolution of the ribosome from an ancient self-replicating machine into its current form as a translational machine may have been the selective pressure to incorporate proteins into the ribosome's self-replicating mechanisms, so as to increase its capacity for self-replication

The RNA world

RNA as catalyst

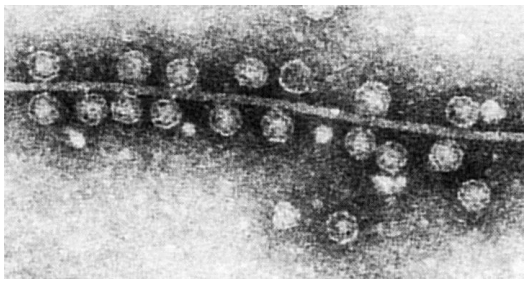
Currently known co-enzymes
Ribozymes
Ribosome

Can RNA evolve?

Can RNA replicate itself?

The RNA world

Can RNA evolve?

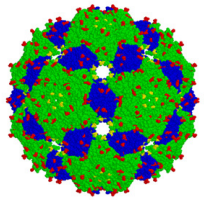


Spiegelman's monster

The RNA world

The bacteriophage Qβ – a virus containing RNA-dependent RNA polymerase (protein, enzymatic replicase)

Spiegelman's monster



Spiegelman mixed the Qβ RNA, the Qβ enzymatic replicase, mononucleotides and some salts (buffer). RNA replication begun.
An aliquot was transferred several times to a fresh solution without template.

Shorter RNA chains replicate faster. The selection in this system favors speed.
And no evolutionary pressure on pathogenicity was present anymore.
So the RNA became shorter and shorter due to random mutations during copying.

After 74 passages, the original 4500 nt RNA strand was reduced to 218 nt.
Such a short RNA chain replicated very quickly under these unnatural circumstances.
Of course, it lost all its genes and was unable to produce any useful proteins anymore.
First example of *in vitro* RNA evolution

Kacian D. L., Mills D. R., Kramer F. R., Spiegelman S. *PNAS* **1972**, *69*, 3038-3042.

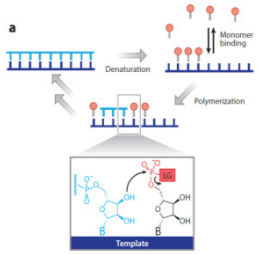
Spiegelman's monster can be also formed by simple mixing of activated RNA monoers and the Qβ enzymatic replicase, in absence of any RNA template!

Sumper M., Luce R. *PNAS* **1975**, *72*, 162-166.

The RNA world

RNA self-replication

Nonenzymatic template-directed RNA polymerization
Maximally 30-50 nt extension, fidelity strongly sequence-dependent

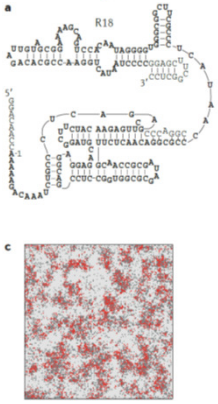


General RNA polymerase ribozyme (‚replicase')

Networks of RNA molecules that mutually catalyse their replication – autocatalytic replication of the whole network

The RNA world

RNA-dependent RNA polymerase ribozyme – Replicase - the ‚holy Grail' of the RNA world



R18 – an artificial polymerase evolved from the class I ligase ribozyme.

Template: another copy of itself (red) or an unrelated sequence (grey).

A sequence of 206 nt was copied (fidelity 97.4%) at low temperatures by an engineered R18 mutant – first ribozyme capable to synthesize RNA oligomers longer than itself (though NO self-replication yet!)

Rate of replication not sensitive on the template's sequence. Replicase could replicate other ribozymes (e.g. with metabolic functions). Self-amplifying replicase needs a working complementary replicase – danger of paraistes (templates that copy themselves but do not contribute to the replication of the polymerase).

Systems of altruistic replicators are destroyed by parasites (grey). Replicators (red) can survive e.g. by diffusion on 2D surfaces (**c**) or selection inside compartments (**d**)

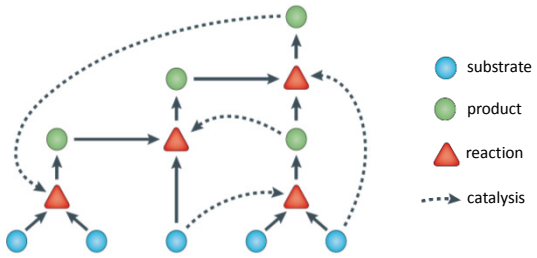
Johnston, W. K., Unrau, P. J., Lawrence, M. S., Glasner, M. E. & Bartel, D. P. *Science* **2001**, *292*, 1319–1325.
Attwater, J., Wochner, A. & Holliger, P. *Nature Chem.* **2013**, *5*, 1011–1018.

The RNA world

Replicase - problem

The replicase most likely needs to be long (> 200 nt) for the efficient replication –
How could such long fucntional RNA be spontaneously generated?

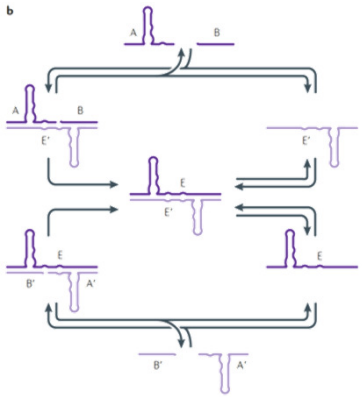
Possible solution – autocatalytic networks



No component can replicate without all the others

The RNA world

Mutually autocatalytic RNA networks

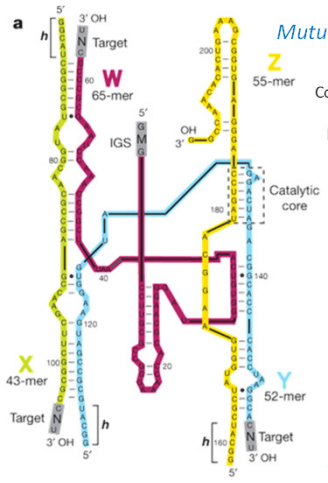


An autocatalytic set composed of two cross-catalytic ligases was demonstrated. RNA A and RNA B are ligated together by ribozyme E' to create ribozyme E, which can reciprocate and ligate RNA A' and RNA B' to create ribozyme E'.

Lincoln, T. A. & Joyce, G. F. *Science* **2009**, *323*, 1229–1232.

The RNA world

Mutually autocatalytic RNA networks



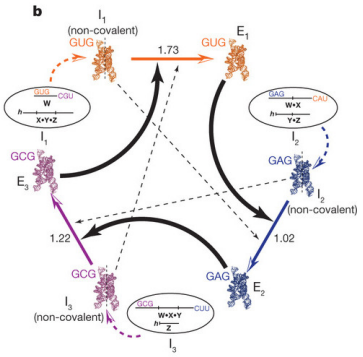
Cooperation between multiple strands that assemble to perform a single function.

Ribozymes, such as the *Azoarcus* recombinase, can be made from several short strands that assemble as a result of RNA secondary structure formation and information contained in internal guide sequences (IGSs) and complementary targets (grey).

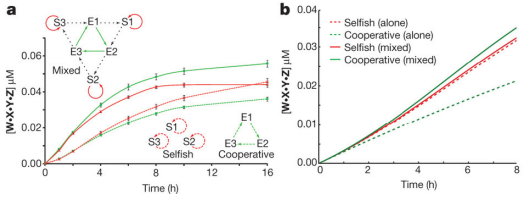
Vadia, N. *et al. Nature* **2012**, *491*, 72-77.

The RNA world

Mutually autocatalytic RNA networks

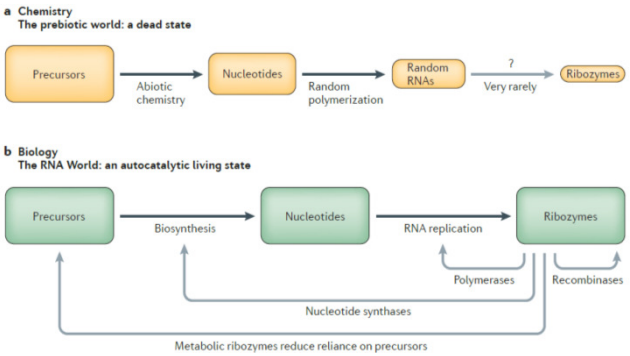


mixtures of RNA fragments that self-assemble into self-replicating ribozymes spontaneously form cooperative catalytic cycles and networks.



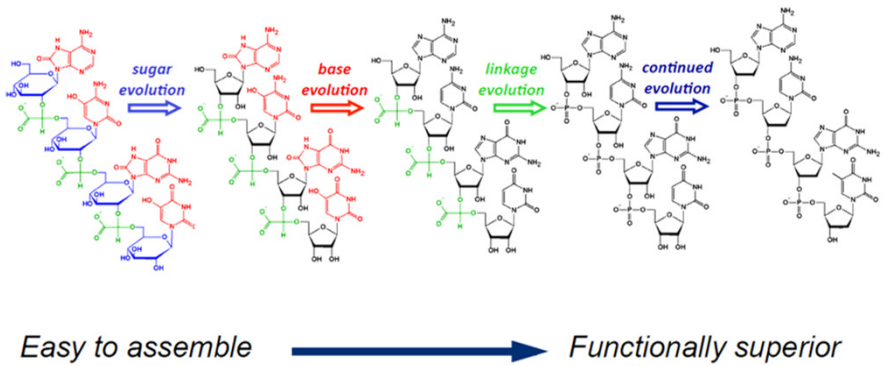
Vadia, N. *et al. Nature* **2012**, *491*, 72-77.

The RNA world



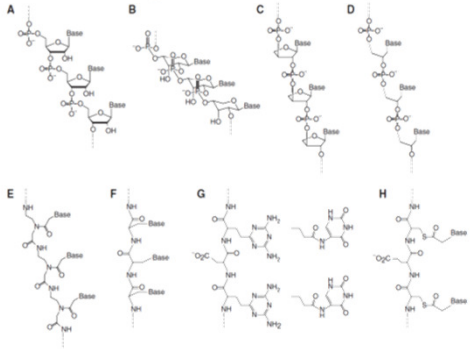
Transition from chemistry to biology involves autocatalytic feedbacks from ribozymes to all stages of the prebiotic chemistry

„RNA-second"



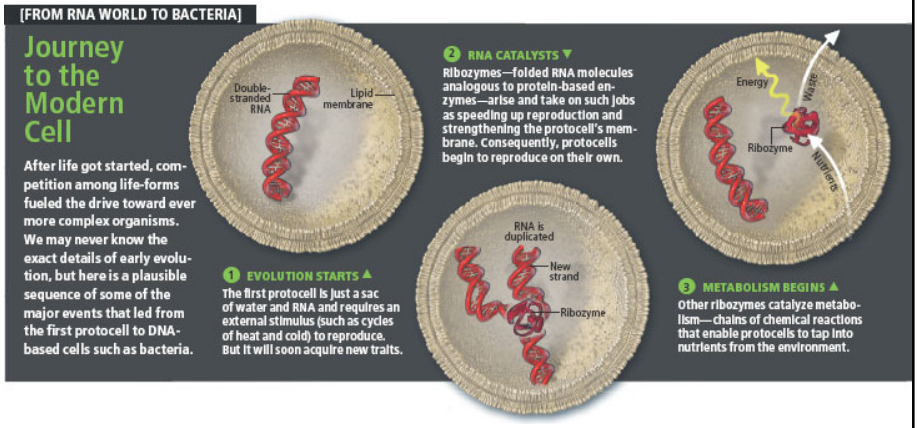
Proto-RNA evolution: According to the protoRNA theory, each of the components of RNA — sugar, base and phosphate backbone — may have originally taken different forms.

„RNA-second"



The structures of (A) RNA; (B) p-RNA; (C) TNA; (D) GNA; (E) PNA; (F) ANA; (G) diaminotriazinetagged (left) and dioxo-5-aminopyrimidine-tagged (right) oligodipeptides; and (H) tPNA. ANA contains a backbone of alternating D- and L-alanine subunits. The diaminotriazine tags are shown linked to a backbone of alternating L-aspartate and L-glutamate subunits; the dioxo-5-aminopyrimidine tags (shown unattached) can be linked similarly. tPNA is shown with a backbone of alternating L-cysteine and L-glutamate subunits.

From RNA world to bacteria



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