Transition from the RNA world to LUCA

Ribozymes – self-acting → metabolic

Evolution of ribosome

Incorporation of aminoacids and peptides

The genetic code and archival storage

Enzyme-driven metabolism and membranes

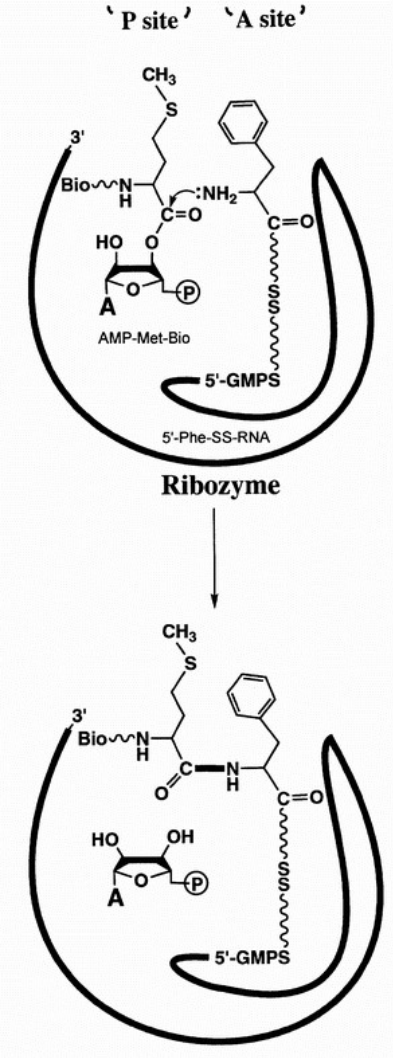
Ribozymes

Initially only self-processing ribozymes (introns, RNAses) discovered.

1992 – first ribozyme isolated capable to cleave the bond of methionine with its tRNA (also the reverse reaction – transacylation – is catalysed)

1995 (Yarus) – a random RNA sequence found capable of attaching an activated aminoacid to itself

1997 (Szostak) – an RNA sequence that transfers one aminoacid to another one, forming a dipeptide → analogue of the peptidetransferase center of the ribosome



Ribozymes

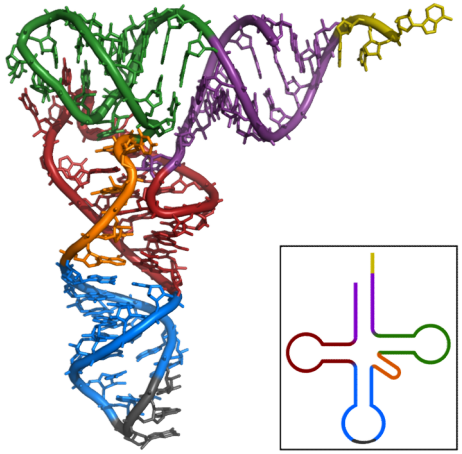
Ribozymes incorporate aminoacids to enhance their catalytic abilities

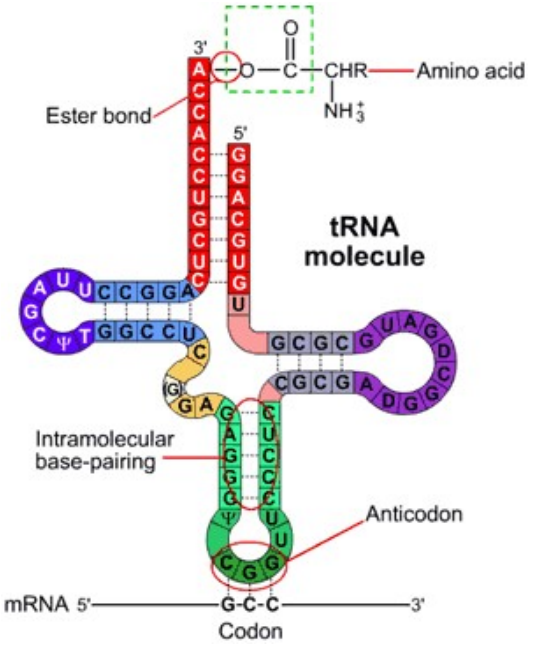
It opens ways to improved metabolism and provides evolutionary advantage in receiving energy from outside

Initially incorporation of aminoacids may have improved synthesis of nucleotides to produce more RNA

Primordial tRNAs were most likely self-charging, today special enzymes do it (tRNA synthethases)

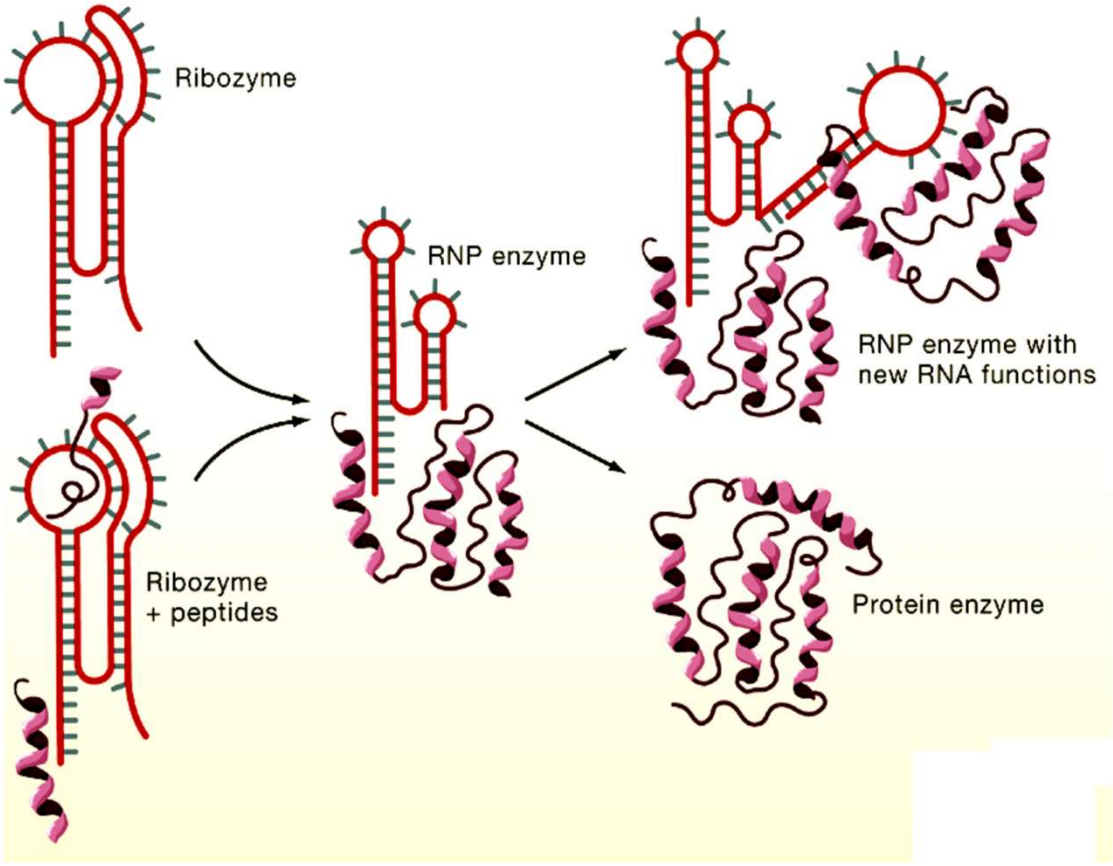
Peptide chains increase in size, the RNA part decrease → non-covalent binding of nucleoside cofactors to contemporary enzymes





aminoacyl-tRNA

Evolution of biocatalysis



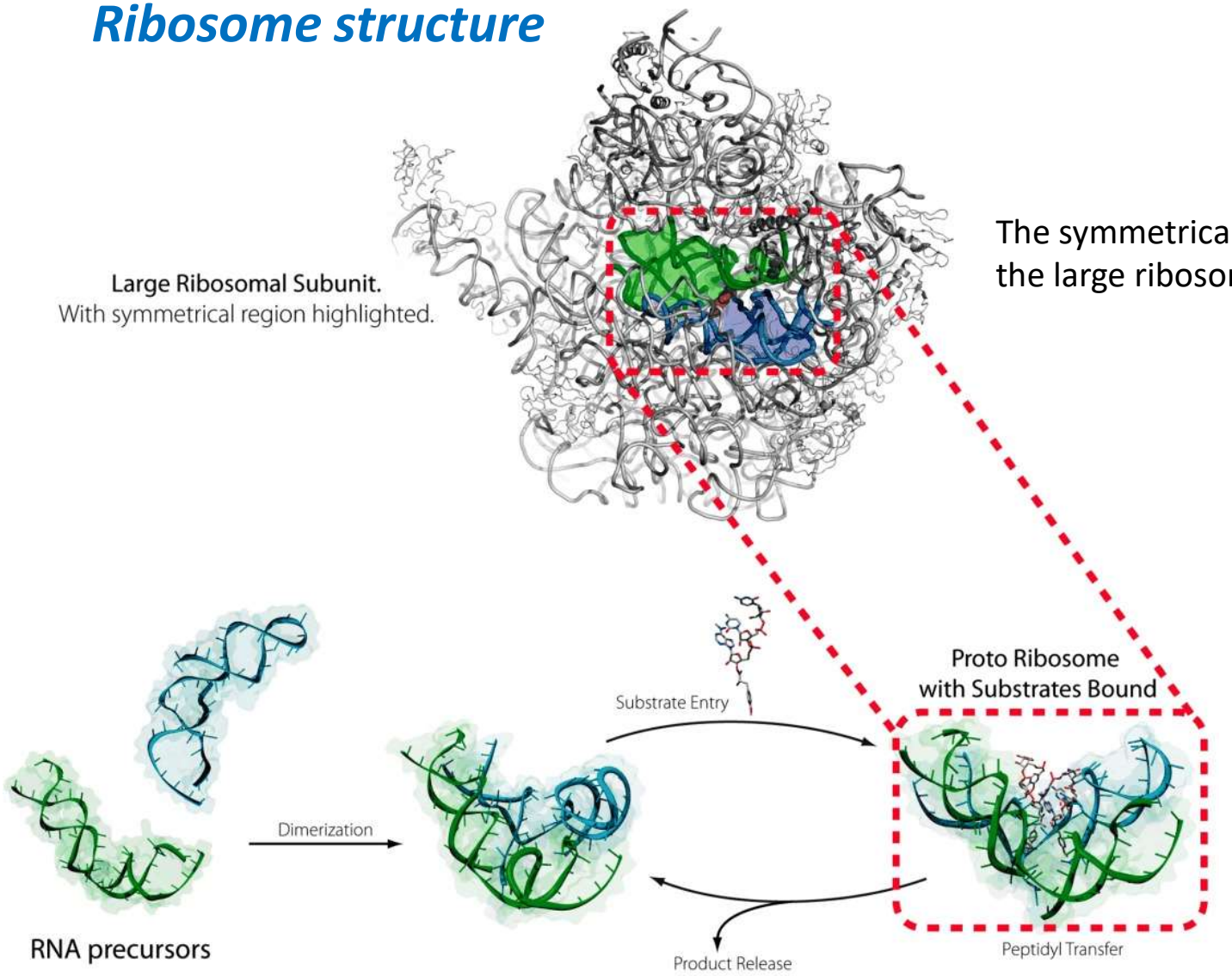
Primordial **RNA-only ribozymes** as well as **complexes of ribozymes and random peptides** could have acted as catalysts during the first steps of the RNA world.

In a more advanced stage, upon the advent of *peptidyltransferase ribozymes*, the availability of RNA-coded proteins allowed the assembly of **ribonucleoprotein (RNP) complexes**.

Some of the RNPs could have shown novel or improved catalytic activities, ultimately including the *translation of mRNA on protoribosomes*. Later, some **RNP enzymes** (upper right) evolved by adding or discarding some RNA subunits and fine-tuning their catalytic activity. In parallel, most RNP complexes (lower right) evolved to **protein-only enzymes**.

Cech, T. R. Cell **2009**, *136*, 599.

Ribosome structure

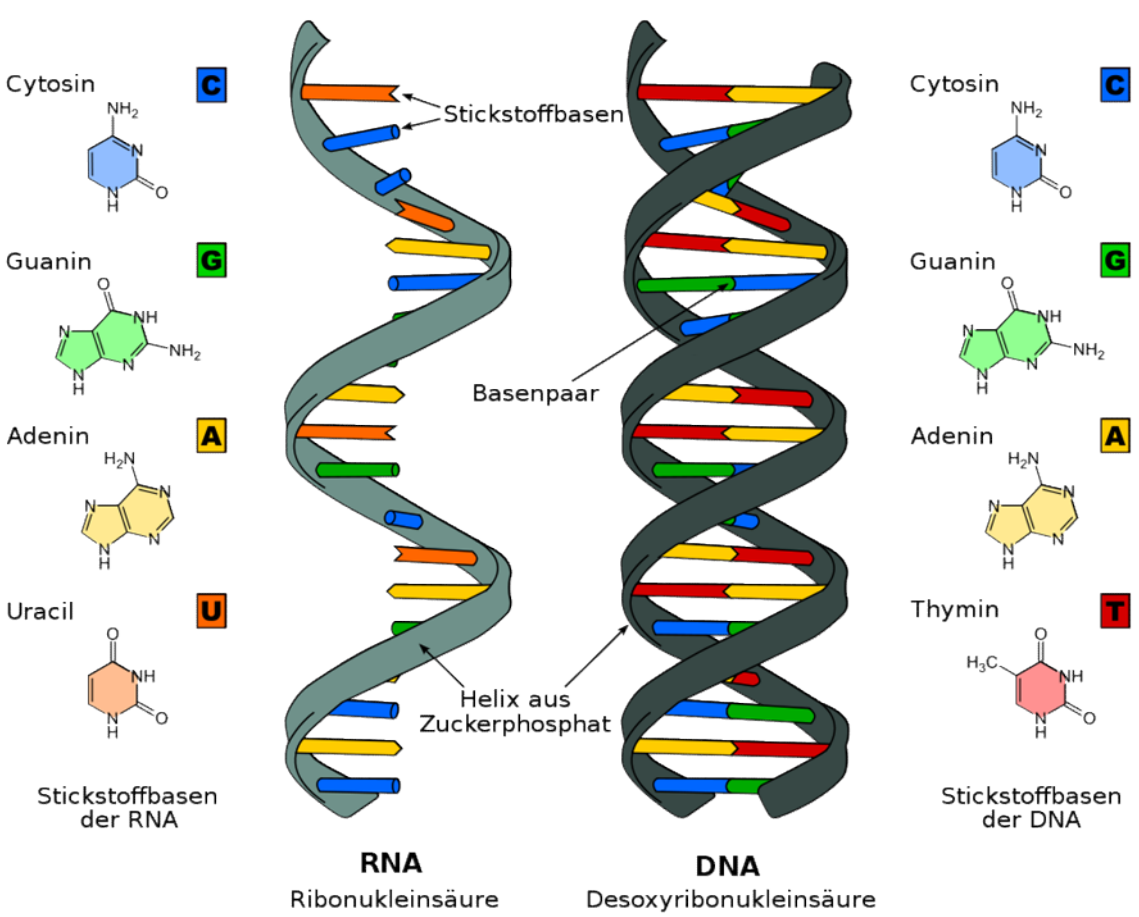


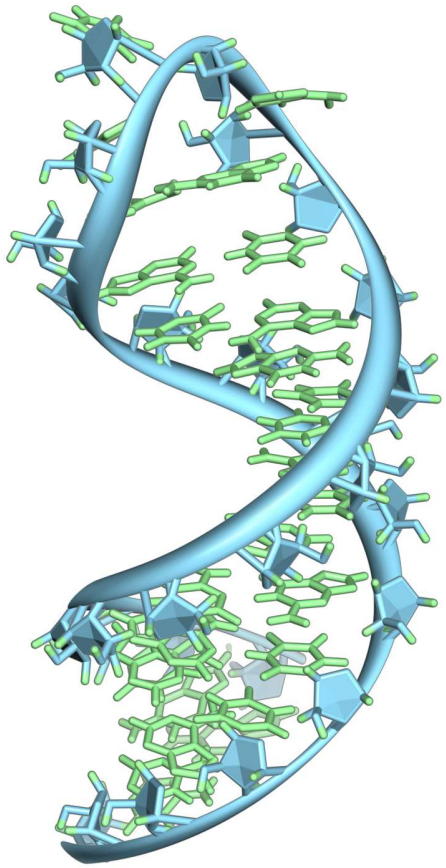
The symmetrical region within the large ribosomal subunit.

Schematic representation of "pocket-like" proto-ribosome formation from a RNA precursor, showing simple catalytic peptidyl transferase activity.

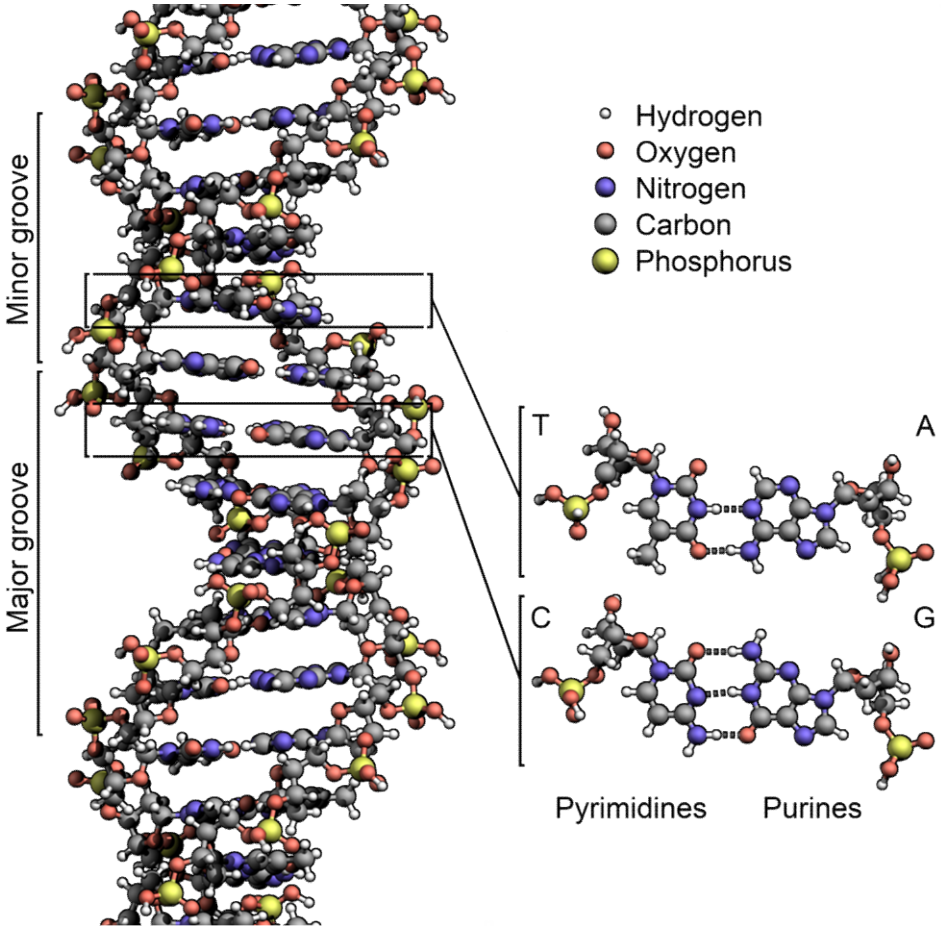
A. Yonath *et al. Israel Journal of Chemistry,* **2010***, 50,* 29-35

The origin of DNA





The origin of DNA



Maximal size of RNA-based genome: 3000-5000 bases
(HIV, West Nile virus)

Reason: above that, statistically certain to generate at least one self-cleaving RNA sequence (ribozyme)

Maximal DNA size – unlimited

- no self-cleaving DNAzymes,
- tight storage as dsDNA,
- methylated uracil (thymine) → no accidental C-to-U mutations

The genetic code

ΔF508 deletion in cystic fibrosis

1st base	2nd base: U	2nd base: C	2nd base: A	2nd base: G
U	UUU (Phe/F) Phenylalanine	UCU (Ser/S) Serine	UAU (Tyr/Y) Tyrosine	UGU (Cys/C) Cysteine
	UUC (Phe/F) Phenylalanine	UCC (Ser/S) Serine	UAC (Tyr/Y) Tyrosine	UGC (Cys/C) Cysteine
	UUA (Leu/L) Leucine	UCA (Ser/S) Serine	UAA Ochre (Stop)	UGA Opal (Stop)
	UUG (Leu/L) Leucine	UCG (Ser/S) Serine	UAG Amber (Stop)	UGG (Trp/W) Tryptophan
C	CUU (Leu/L) Leucine	CCU (Pro/P) Proline	CAU (His/H) Histidine	CGU (Arg/R) Arginine
	CUC (Leu/L) Leucine	CCC (Pro/P) Proline	CAC (His/H) Histidine	CGC (Arg/R) Arginine
	CUA (Leu/L) Leucine	CCA (Pro/P) Proline	CAA (Gln/Q) Glutamine	CGA (Arg/R) Arginine
	CUG (Leu/L) Leucine	CCG (Pro/P) Proline	CAG (Gln/Q) Glutamine	CGG (Arg/R) Arginine
A	AUU (Ile/I) Isoleucine	ACU (Thr/T) Threonine	AAU (Asn/N) Asparagine	AGU (Ser/S) Serine
	AUC (Ile/I) Isoleucine	ACC (Thr/T) Threonine	AAC (Asn/N) Asparagine	AGC (Ser/S) Serine
	AUA (Ile/I) Isoleucine	ACA (Thr/T) Threonine	AAA (Lys/K) Lysine	AGA (Arg/R) Arginine
	AUG (Met/M) Methionine	ACG (Thr/T) Threonine	AAG (Lys/K) Lysine	AGG (Arg/R) Arginine
G	GUU (Val/V) Valine	GCU (Ala/A) Alanine	GAU (Asp/D) Aspartic acid	GGU (Gly/G) Glycine
	GUC (Val/V) Valine	GCC (Ala/A) Alanine	GAC (Asp/D) Aspartic acid	GGC (Gly/G) Glycine
	GUA (Val/V) Valine	GCA (Ala/A) Alanine	GAA (Glu/E) Glutamic acid	GGA (Gly/G) Glycine
	GUG (Val/V) Valine	GCG (Ala/A) Alanine	GAG (Glu/E) Glutamic acid	GGG (Gly/G) Glycine

3rd base in each row

- Myotonic dystrophy
- SCA 8

β-Thalassemia

McArdle's disease

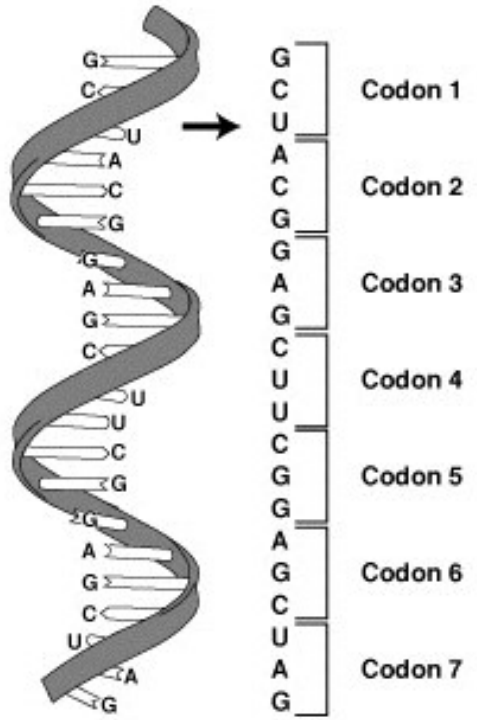
β-Thalassem

Prostate cancer

Colorectal cancer

Sickle-cell disease

Friedreich's ataxia

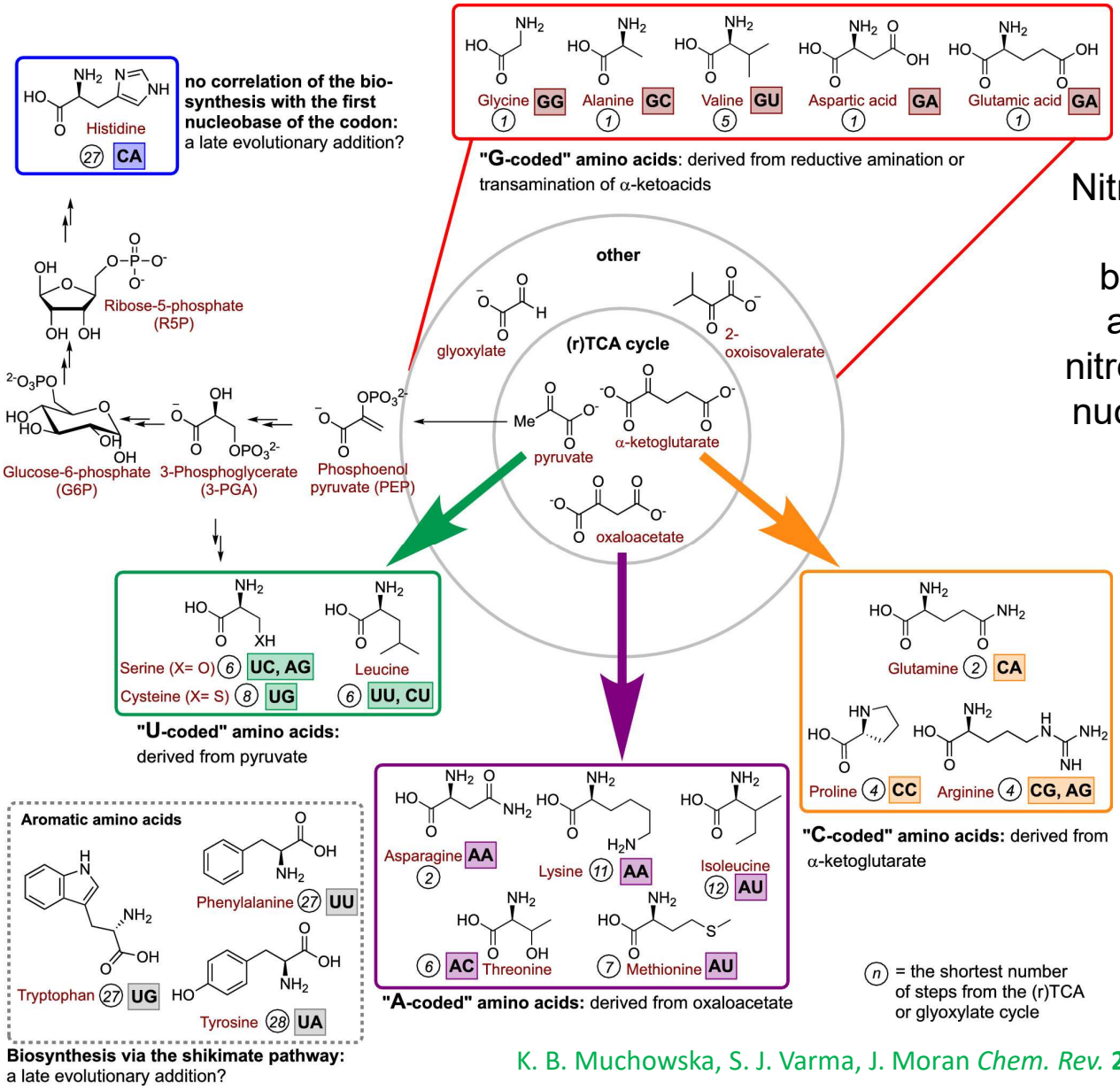


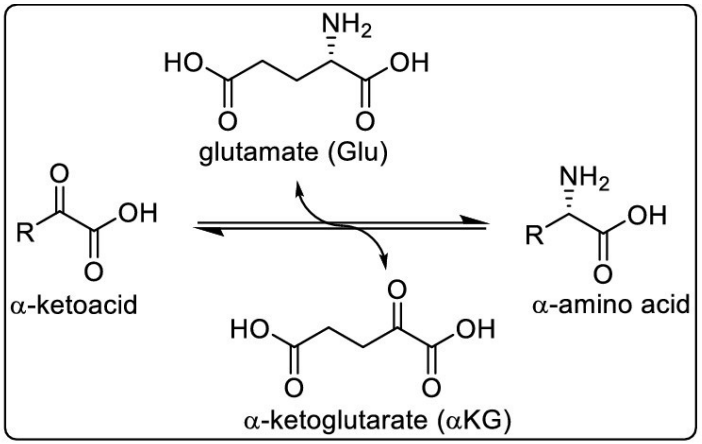
RNA

Ribonucleic acid

The genetic code

Nitrogen enters metabolism almost exclusively via two aminoacids. **Glutamine** is biosynthesized when glutamate reacts with ammonia and ATP and serves mainly as a nitrogen donor in the biosynthesis of pyrimidine nucleobases. The main role of **glutamate** is to transfer nitrogen to α-ketoacids in transamination reactions.

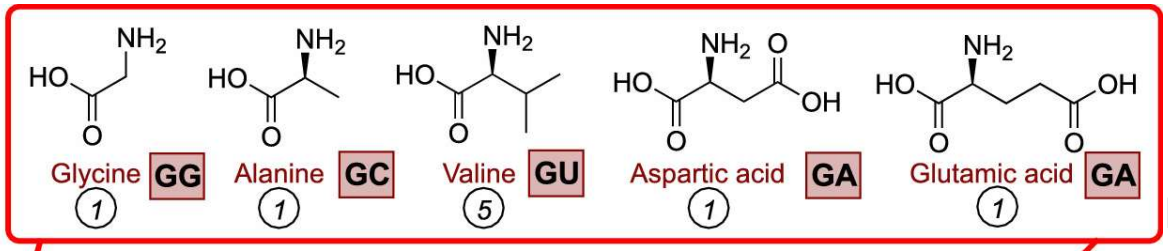




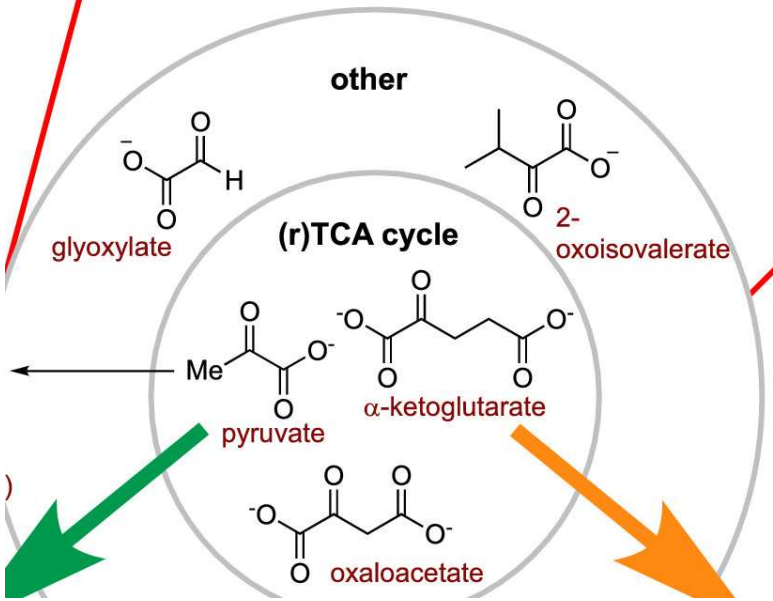
K. B. Muchowska, S. J. Varma, J. Moran *Chem. Rev.* **2020**, *120*, 7708−7744

The genetic code

Glycine, alanine, valine, and aspartate are synthesized directly from their corresponding α-ketoacids. Glutamate can also be synthesized from α-ketoglutarate, by transamination from glutamine.

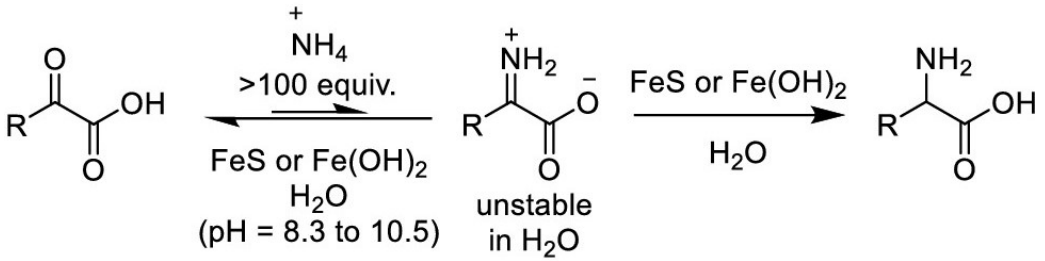


"G-coded" amino acids: derived from reductive amination or transamination of α-ketoacids



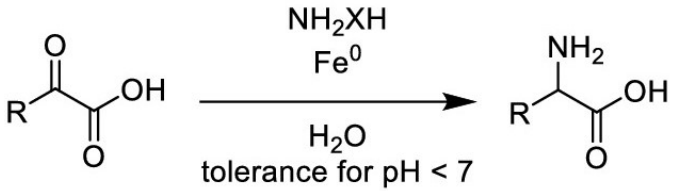
Prebiotically plausible aminations of ketoacids

b) Reductive amination with ammonia



Huber and Wächtershäuser (ref. 221), Barge and co-workers (ref. 222)

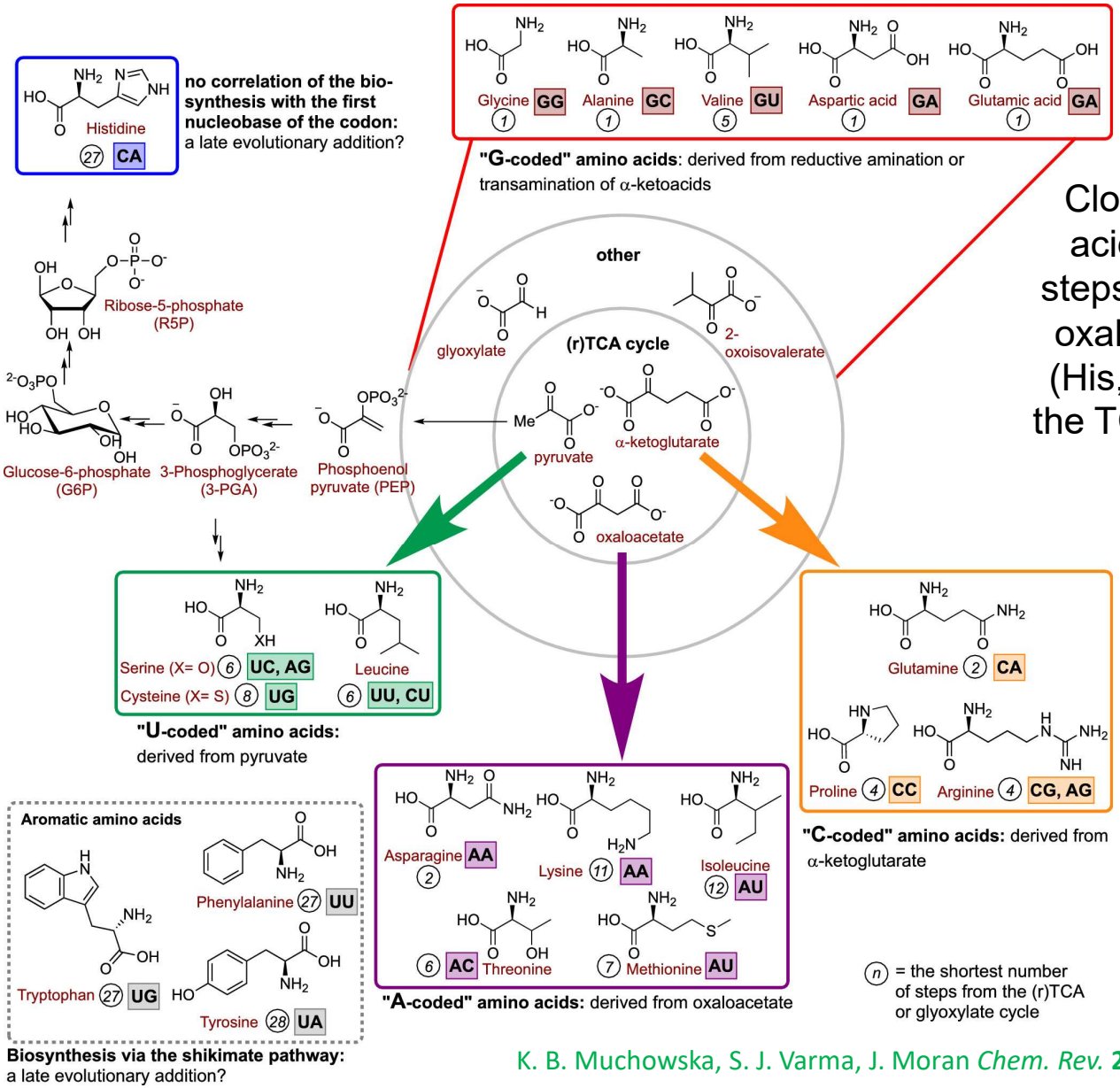
c) Reductive amination with bifunctional amines



K. B. Muchowska, S. J. Varma, J. Moran *Chem. Rev.* **2020**, *120*, 7708−7744

Moran and co-workers (ref. 132, 157)

The genetic code



Close examination of the 20 proteinogenic amino acids reveals that 16 are biosynthesized in 1−12 steps from three TCA cycle intermediates: pyruvate, oxaloacetate, and α-ketoglutarate. The four others (His, Phe, Tyr, Trp) are also ultimately derived from the TCA cycle but are much further away (>20 steps)

If a nonenzymatic (r)TCA cycle, or something that produced the same key ketoacids, was the core of prebiotic chemistry, we might consider the 16 amino acids to be the most ancient, whereas the four synthesized in >20 steps would be later developments.

K. B. Muchowska, S. J. Varma, J. Moran *Chem. Rev.* **2020**, *120*, 7708−7744

The genetic code

The biosynthesis of amino acids bears strong evolutionary ties to the biosynthesis and evolution of genetics that might date back to **prebiotic chemistry**.

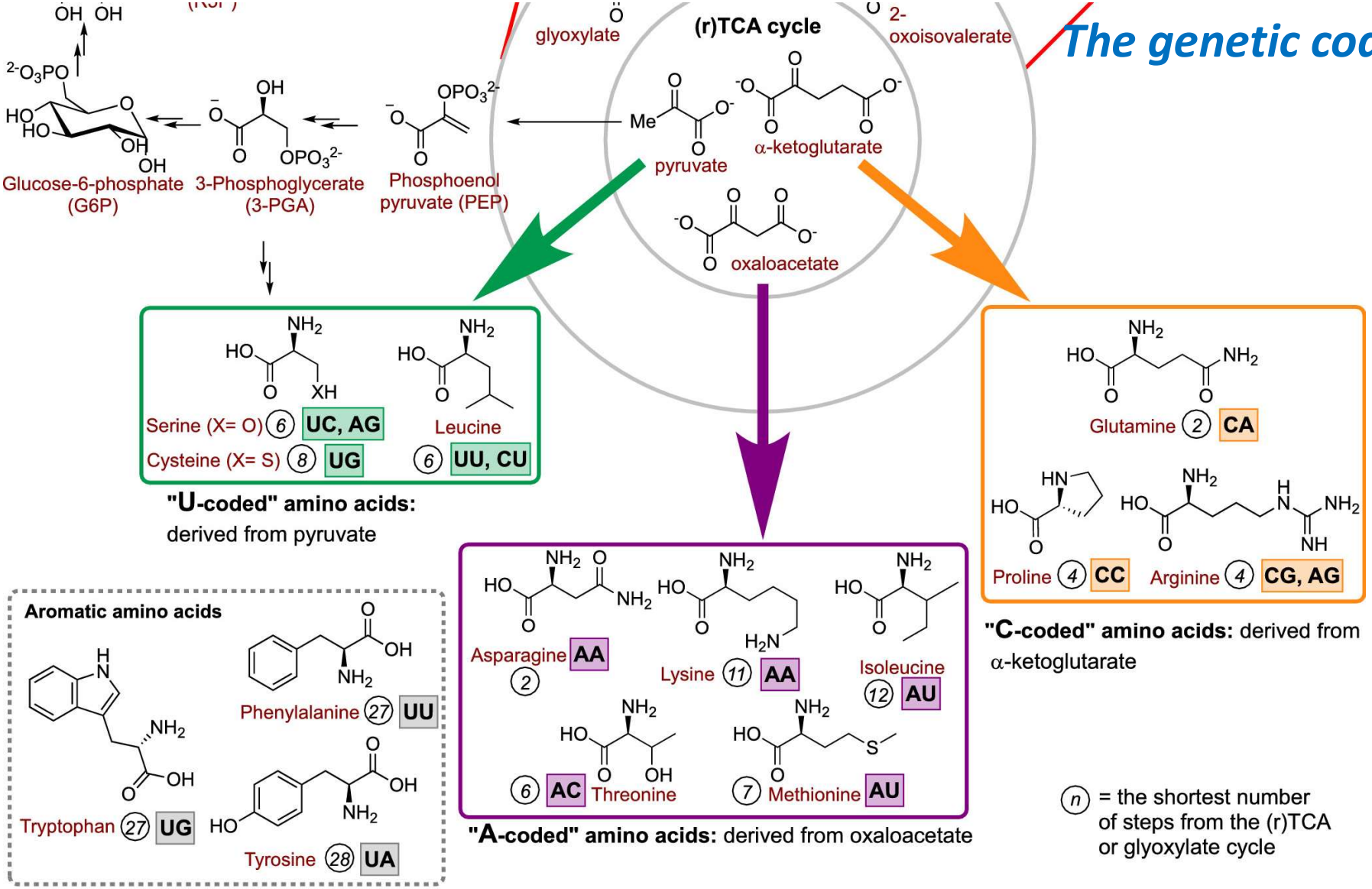
The code contains many unexplained patterns, most notably a strong correlation between the **first two bases** in a codon and **the biosynthetic pathway** used to make the coded amino acid.

The third base of most codons is highly redundant and therefore contains less information than the first two bases. By omitting the third base, a **simplified doublet code** was proposed which can be viewed as a **set of biosynthetic instructions** for the coded amino acid.

Of the 16 “ancient” amino acids, those whose codon starts with cytidine (C) are derived from ketoglutarate, while for adenine (A) it is oxaloacetate, and for uracil (U) it is pyruvate. The second base of the codon is also predictive of the later transformations in amino acid biosynthesis

K. B. Muchowska, S. J. Varma, J. Moran *Chem. Rev.* **2020**, *120*, 7708−7744

The genetic code



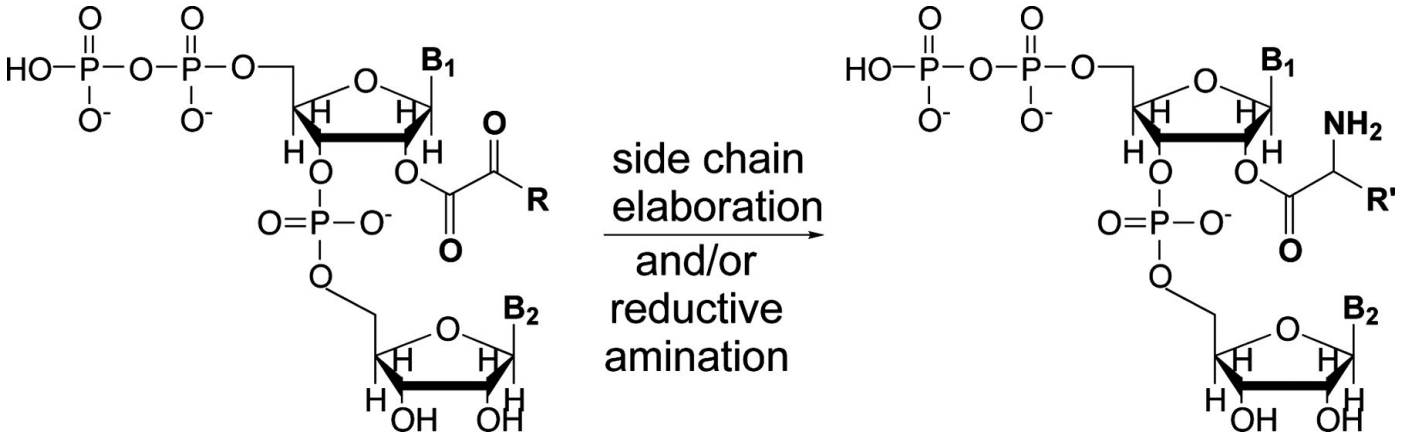
K. B. Muchowska, S. J. Varma, J. Moran *Chem. Rev.* **2020**, *120*, 7708−7744

The genetic code

Why should the code be more strongly correlated with the AA's biosynthetic history than it is with its chemical properties?

An appealing hypothesis is that within a protometabolism, diribonucleotides would become covalently bound to amino acid precursors and induce intramolecular catalysis of specific, sequential steps of amino acid biosynthesis. In other words, amino acids would be made right on the diribonucleotide, producing the associations now found in the genetic code that correlates with the amino acid's synthetic history.

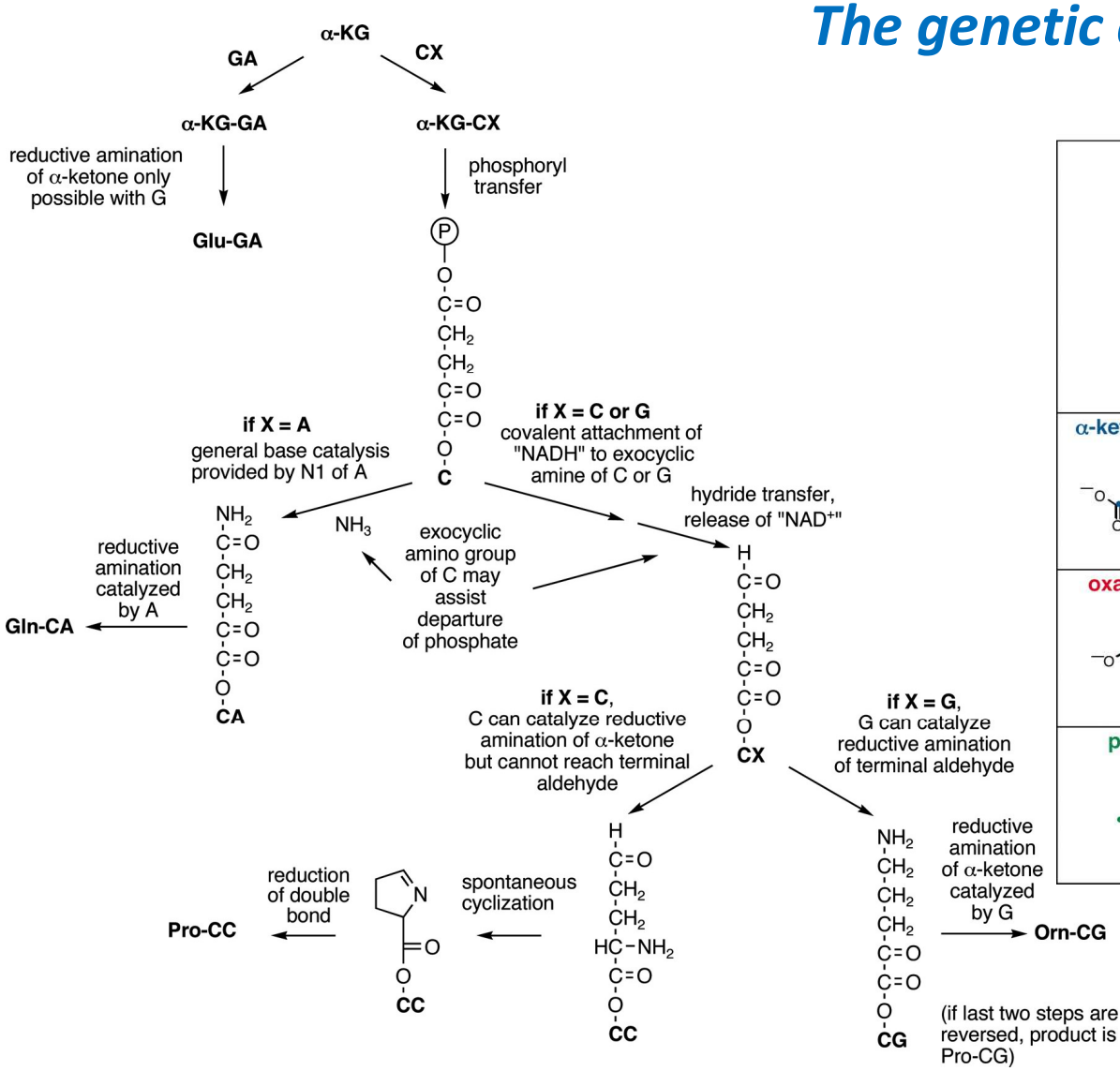
A set of organocatalytic mechanisms for the alleged intramolecular catalysis has been theoretically proposed, but not experimentally verified yet.

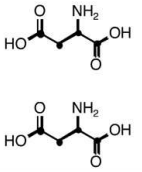
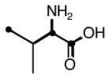
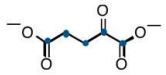
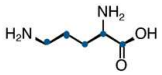
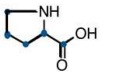
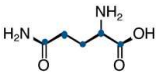
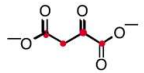
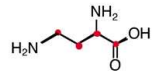
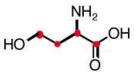
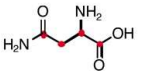
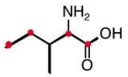
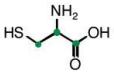
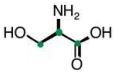
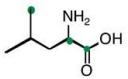


Model for synthesis of amino acids from α-keto acid precursors covalently attached to dinucleotides. The dinucleotide that is capable of catalyzing synthesis of a particular amino acid is proposed to contain the first two bases of the codon specifying that amino acid.

Copley, S. D.; Smith, E.; Morowitz, H. J. *Proc. Natl. Acad. Sci. U. S. A.* **2005**, *102*, 4442−4447

The genetic code

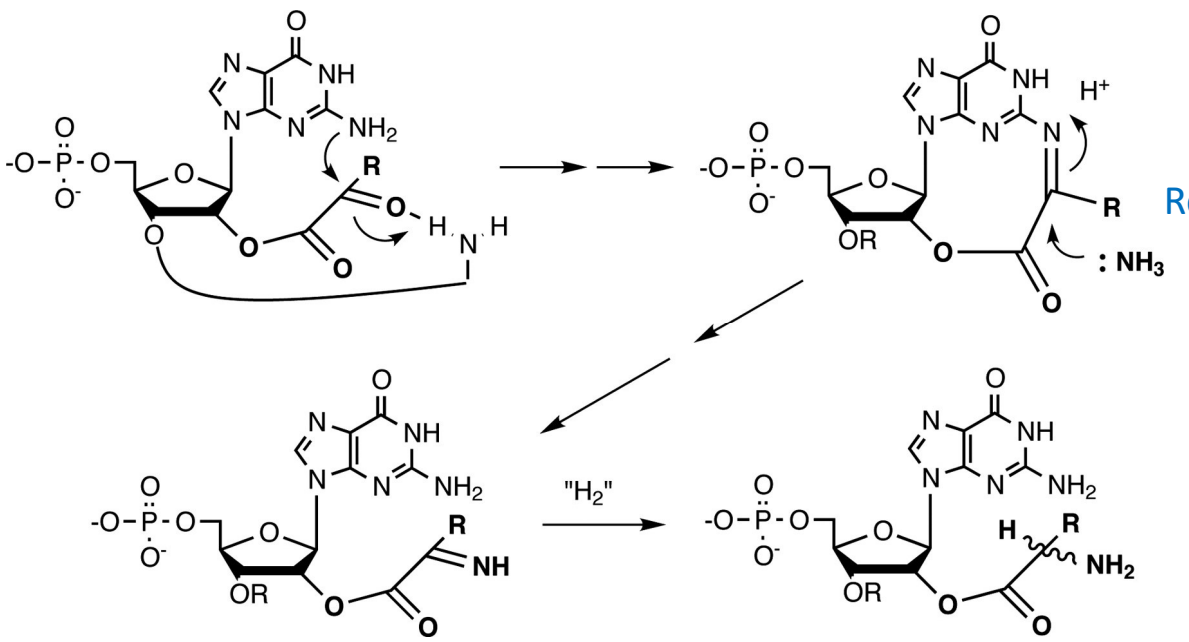


	first position	second position: G	C	A	U
	G	Gly	Ala	Asp/Glu	Val
		Gly	Ala	Asp/Glu	Val
					
α-ketoglutarate 	C	Arg	Pro	Gln	Leu
		Orn	Pro	Gln	?
					?
oxaloacetate 	A	Ser / Arg	Thr	Asn	Ile
		Dab	Hsr	Asn	Ile
					
pyruvate 	U	Cys	Ser	Tyr/stop	Leu
		Cys	Ser	?	Leu
				?	

A simplified doublet genetic code produced from the modern genetic code

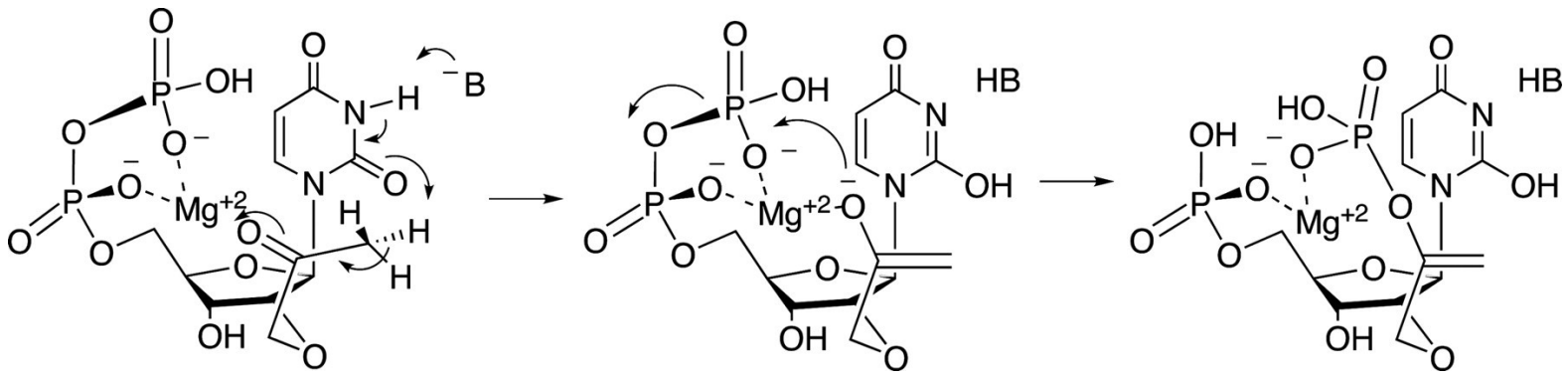
Copley, S. D.; Smith, E.; Morowitz, H. J. *Proc. Natl. Acad. Sci. U. S. A.* **2005**, *102*, 4442−4447

The genetic code



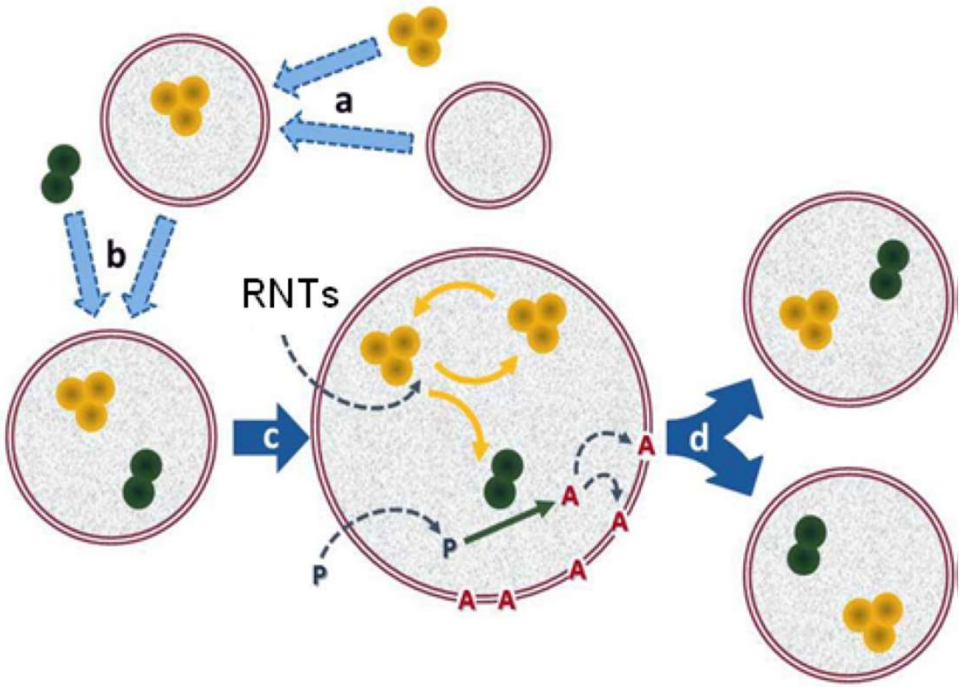
Reductive amination of a covalently attached α-keto acid catalyzed by G in the first position of the dinucleotide catalyst (R the second nucleotide)

Potential mechanism for formation of phosphoenolpyruvate (the first step in the pathway for synthesis of Ser and Cys, as well as many sugars) catalyzed by UpU.



Copley, S. D.; Smith, E.; Morowitz, H. J. *Proc. Natl. Acad. Sci. U. S. A.* **2005**, *102*, 4442−4447

Primordial synthesis of an RNA-based protocell

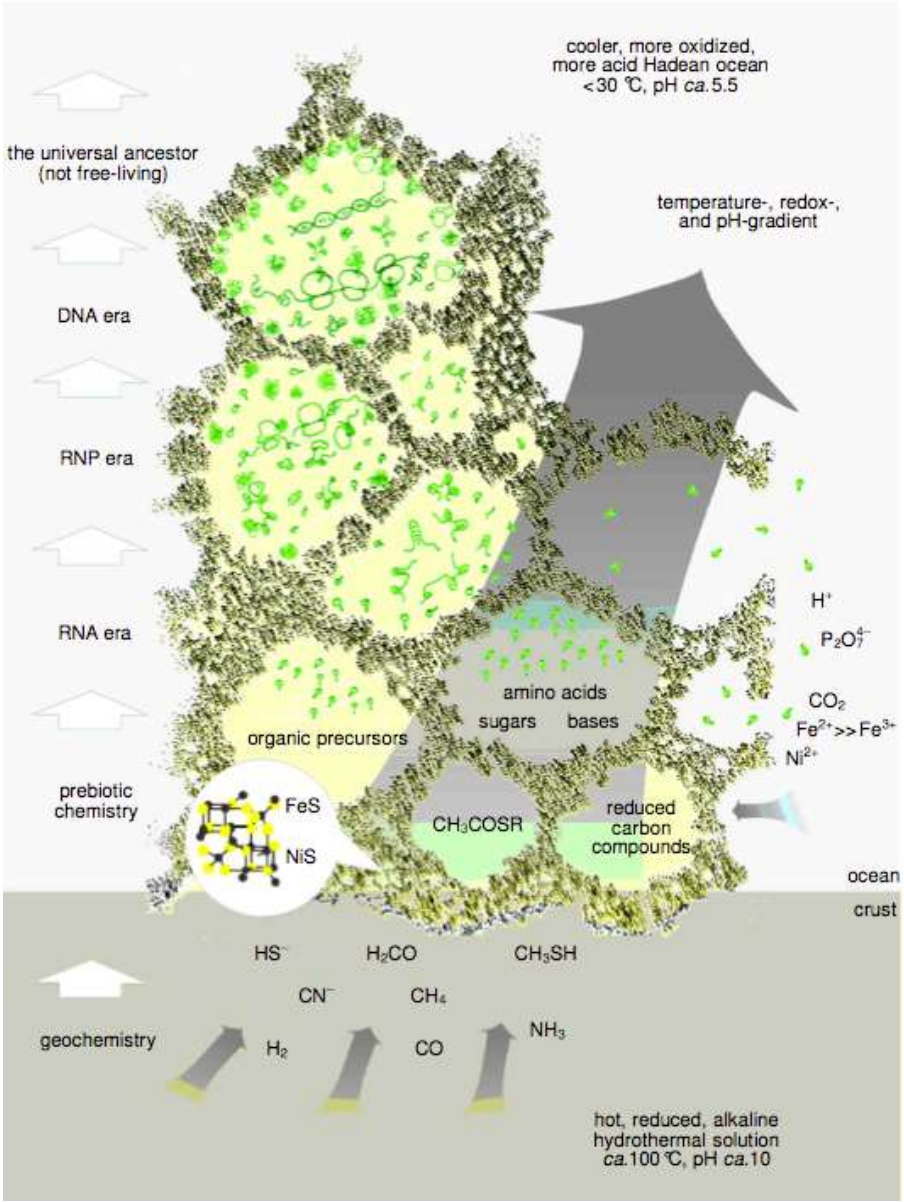


(a) A self-reproducing vesicle is combined with an RNA replicase (yellow).

(b) This system is further combined with a second ribozyme (green) that is able to synthesize amphiphilic molecules (A) from precursor substrates (P), thus leading to an RNA protocell containing two ribozymes.

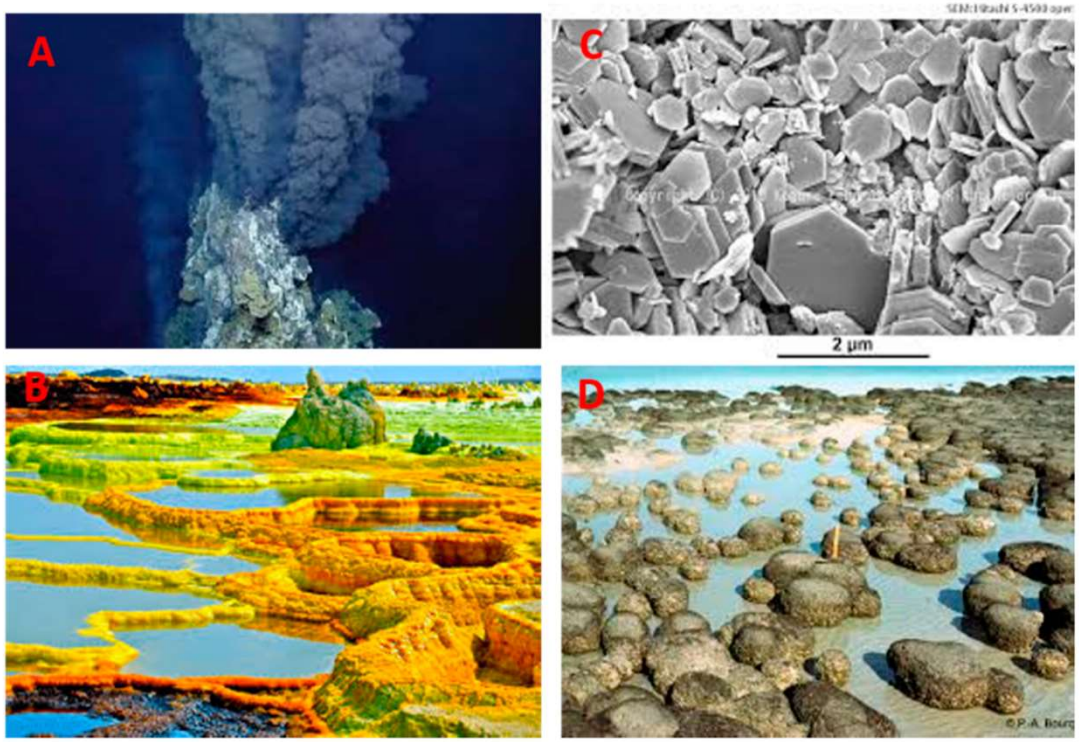
(c) In such a “ribocyte”, the RNA replicase is capable of replicating itself and also making copies of the membrane-forming ribozyme, provided that ribonucleotides (RNTs) are available in the surrounding medium and can permeate the vesicle membrane.

(d) Activity of the second ribozyme converts the previously internalized precursors into amphiphiles, which are further incorporated into the membrane; this leads to a progressive increase of the vesicle size and its subsequent division into two daughter vesicles, thus triggering Darwinian evolution of the whole (membrane−genome coupled) system



Compartments in hydrothermal vents?

Complex metabolic machinery closed in the same compartment that genetic polymers (RNA) which generated it.



A. Lopez, M. Fiore *Life* **2019**, 9(49), doi:10.3390/life9020049

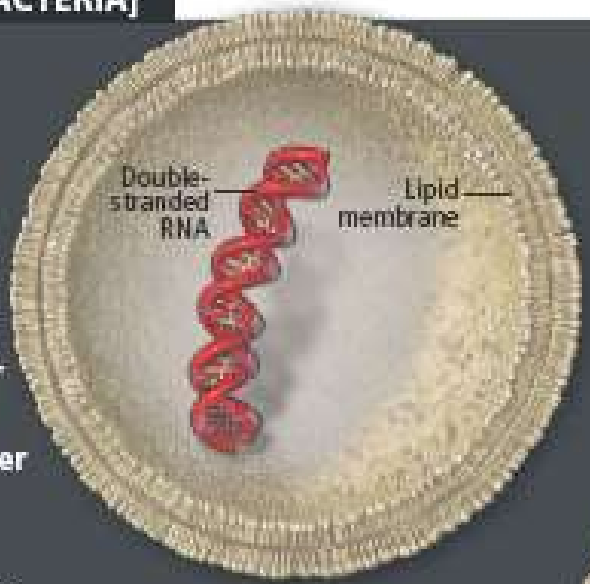
We don't see ribozyme-based metabolism today anymore, because protein catalysts (enzymes) for the same reactions are orders of magnitude faster than the ribozymes

From RNA world to bacteria

[FROM RNA WORLD TO BACTERIA]

Journey to the Modern Cell

After life got started, competition among life-forms fueled the drive toward ever more complex organisms. We may never know the exact details of early evolution, but here is a plausible sequence of some of the major events that led from the first protocell to DNA-based cells such as bacteria.

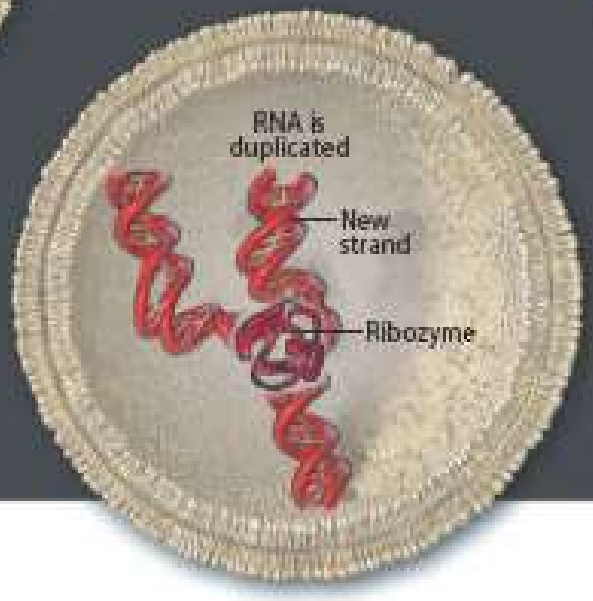


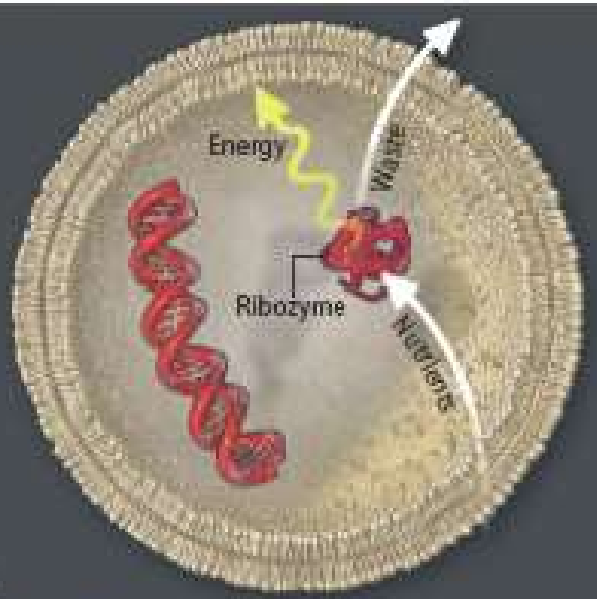
1 EVOLUTION STARTS ▲

The first protocell is just a sac of water and RNA and requires an external stimulus (such as cycles of heat and cold) to reproduce. But it will soon acquire new traits.

2 RNA CATALYSTS ▼

Ribozymes—folded RNA molecules analogous to protein-based enzymes—arise and take on such jobs as speeding up reproduction and strengthening the protocell's membrane. Consequently, protocells begin to reproduce on their own.

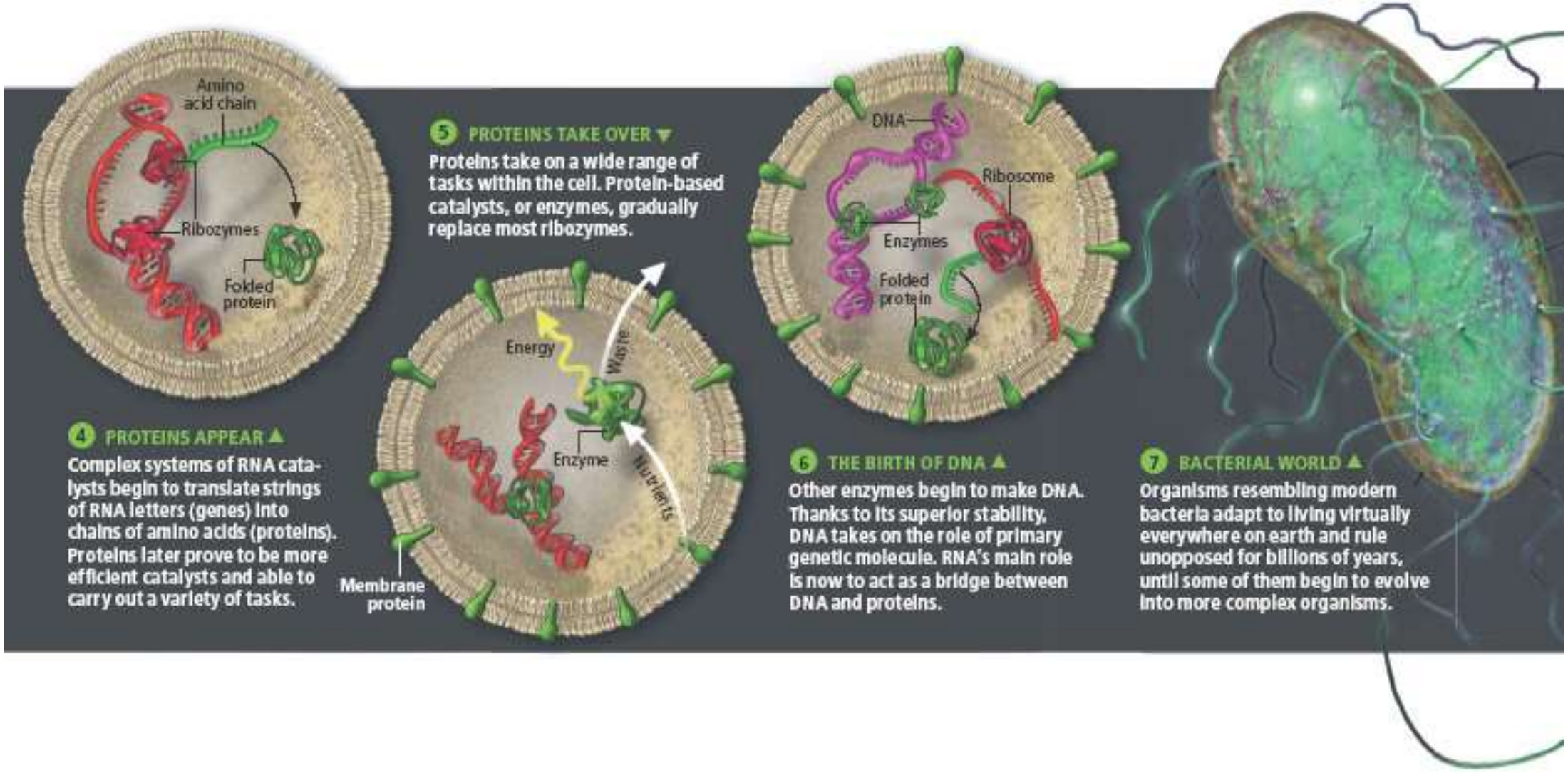




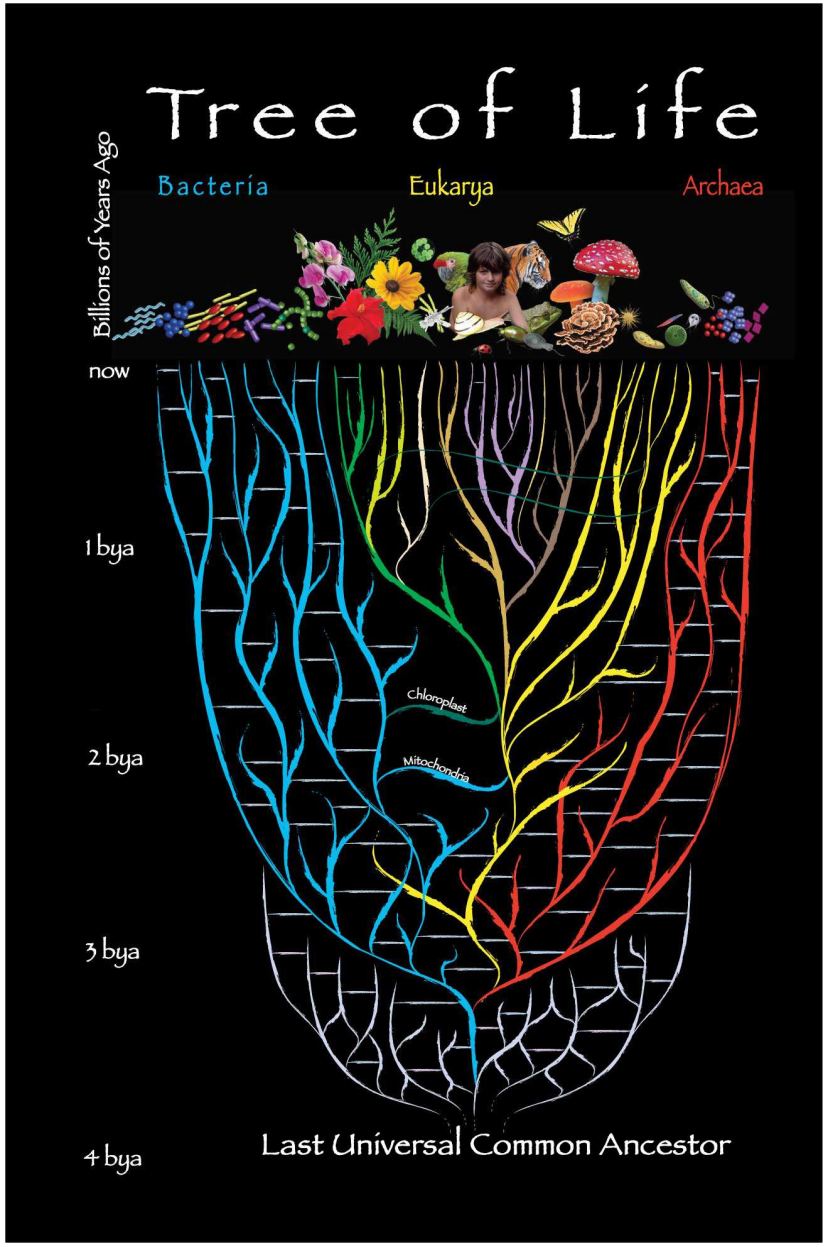
3 METABOLISM BEGINS ▲

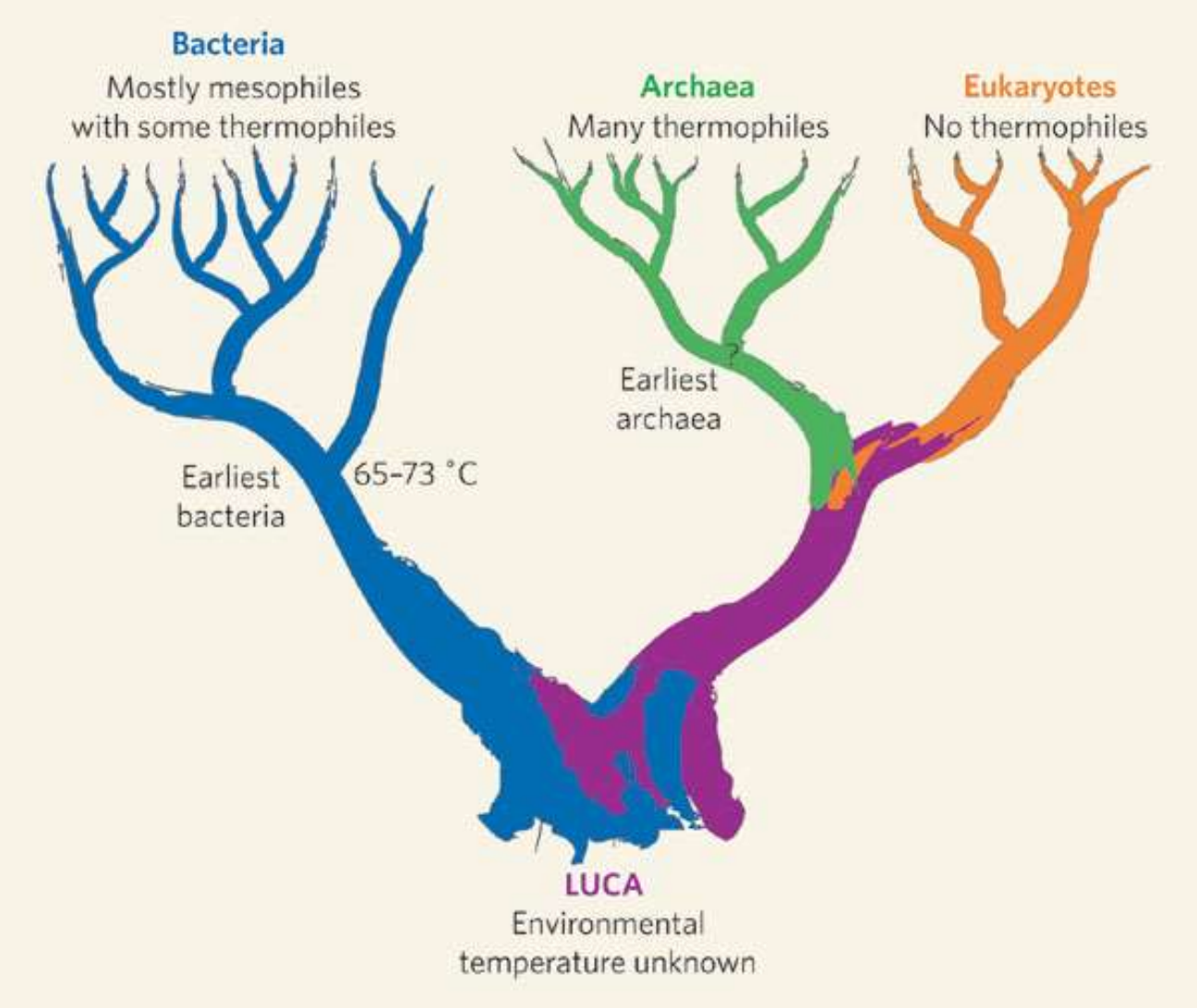
Other ribozymes catalyze metabolism—chains of chemical reactions that enable protocells to tap into nutrients from the environment.

From RNA world to bacteria




LUCA
Last Universal Common Ancestor


Tree of Life
Billions of Years Ago
Bacteria
Eukarya
Archaea
now
1 bya
2 bya
3 bya
4 bya
Chloroplast
Mitochondria
Last Universal Common Ancestor



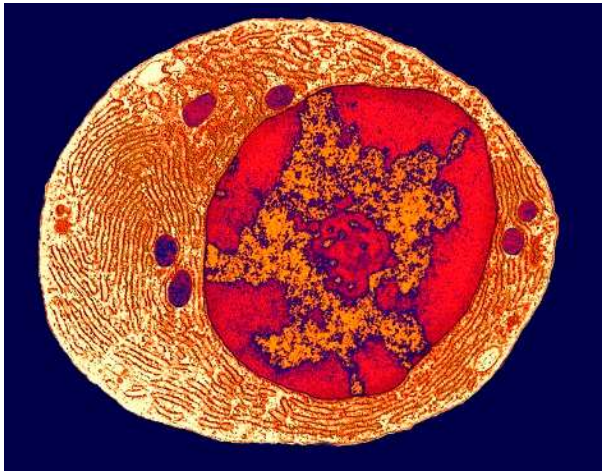
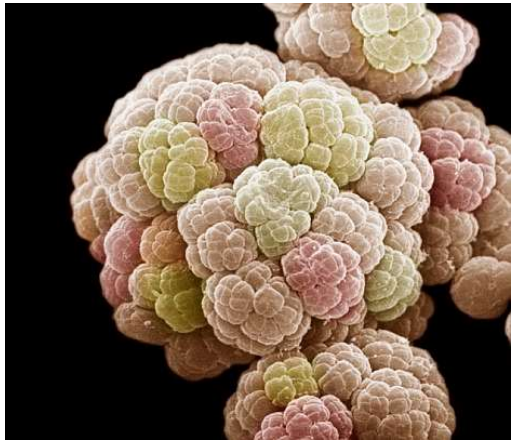


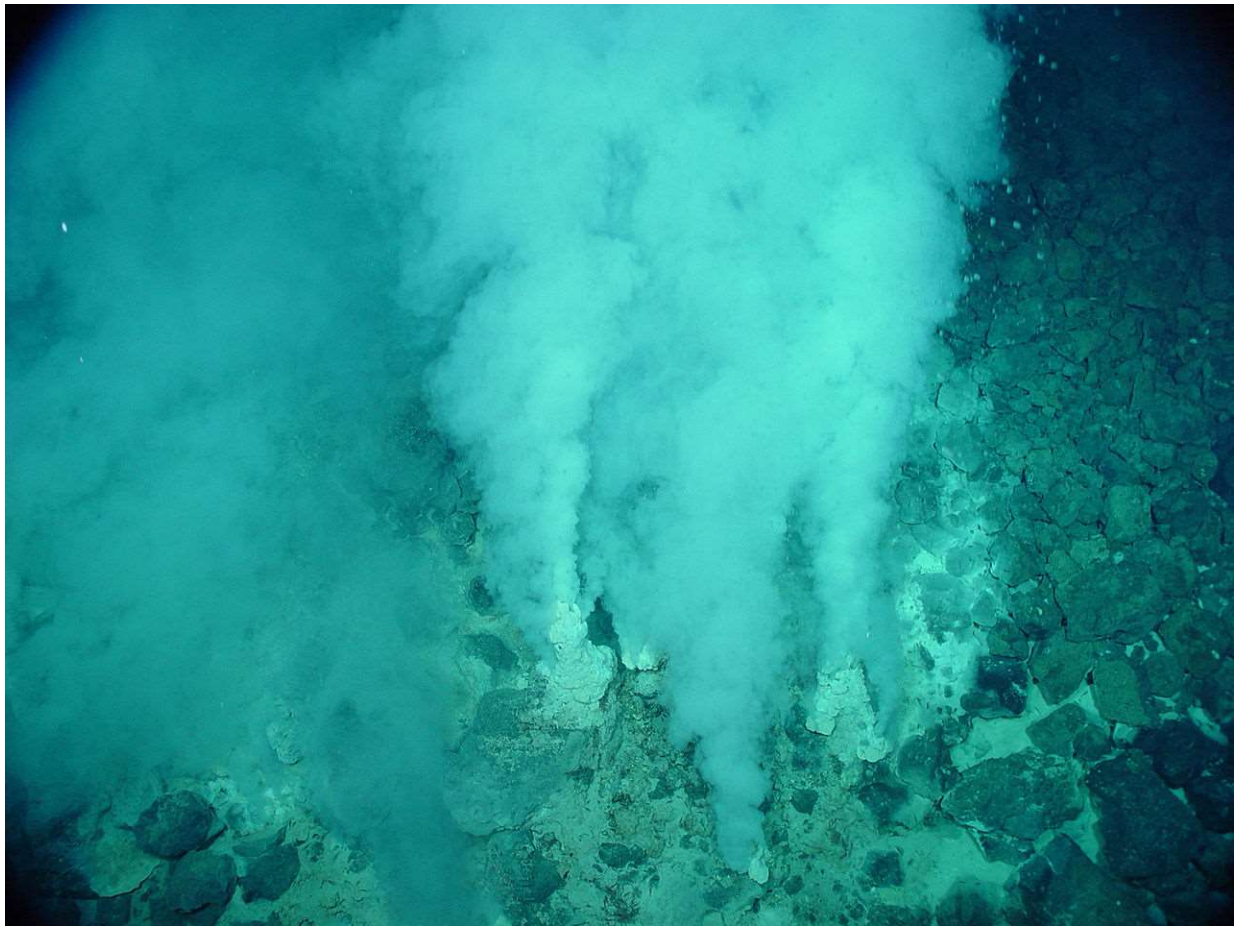
Image of a eukaryotic cell contains numerous organelles, which are now thought to be present in the last universal common ancestor



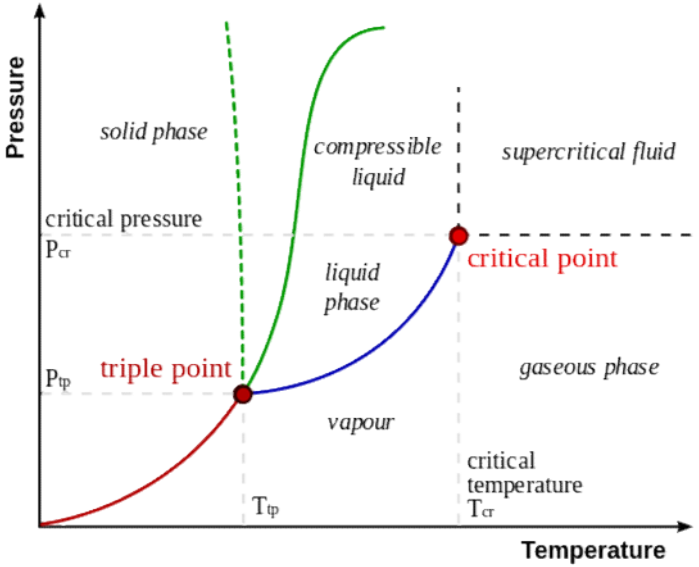
A colony of the archaea, which form one of the three lines of the tree of life in evolutionary history

The Origin of Life near Deep Sea Hydrothermal vents?

Hydrothermal vents

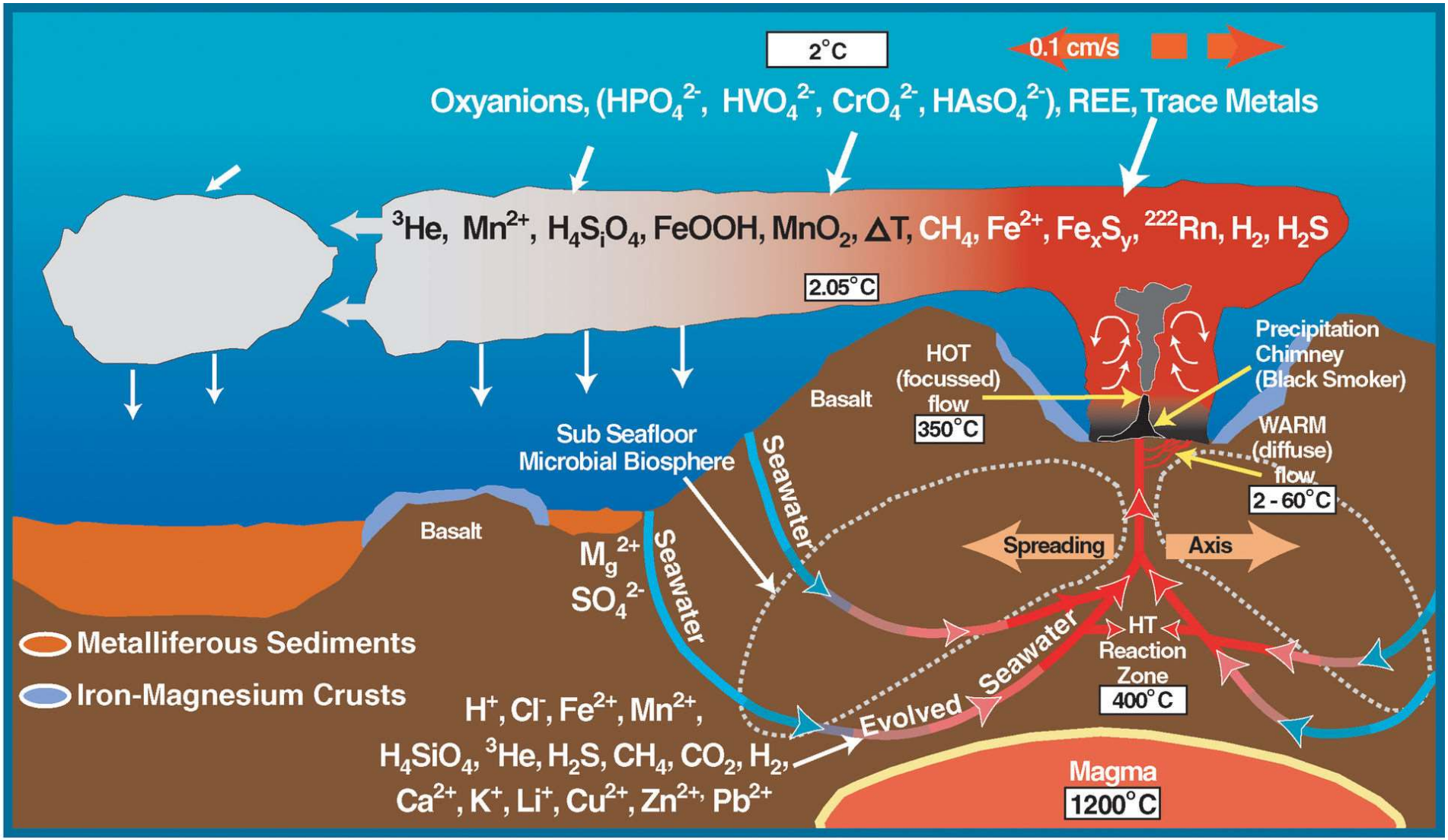


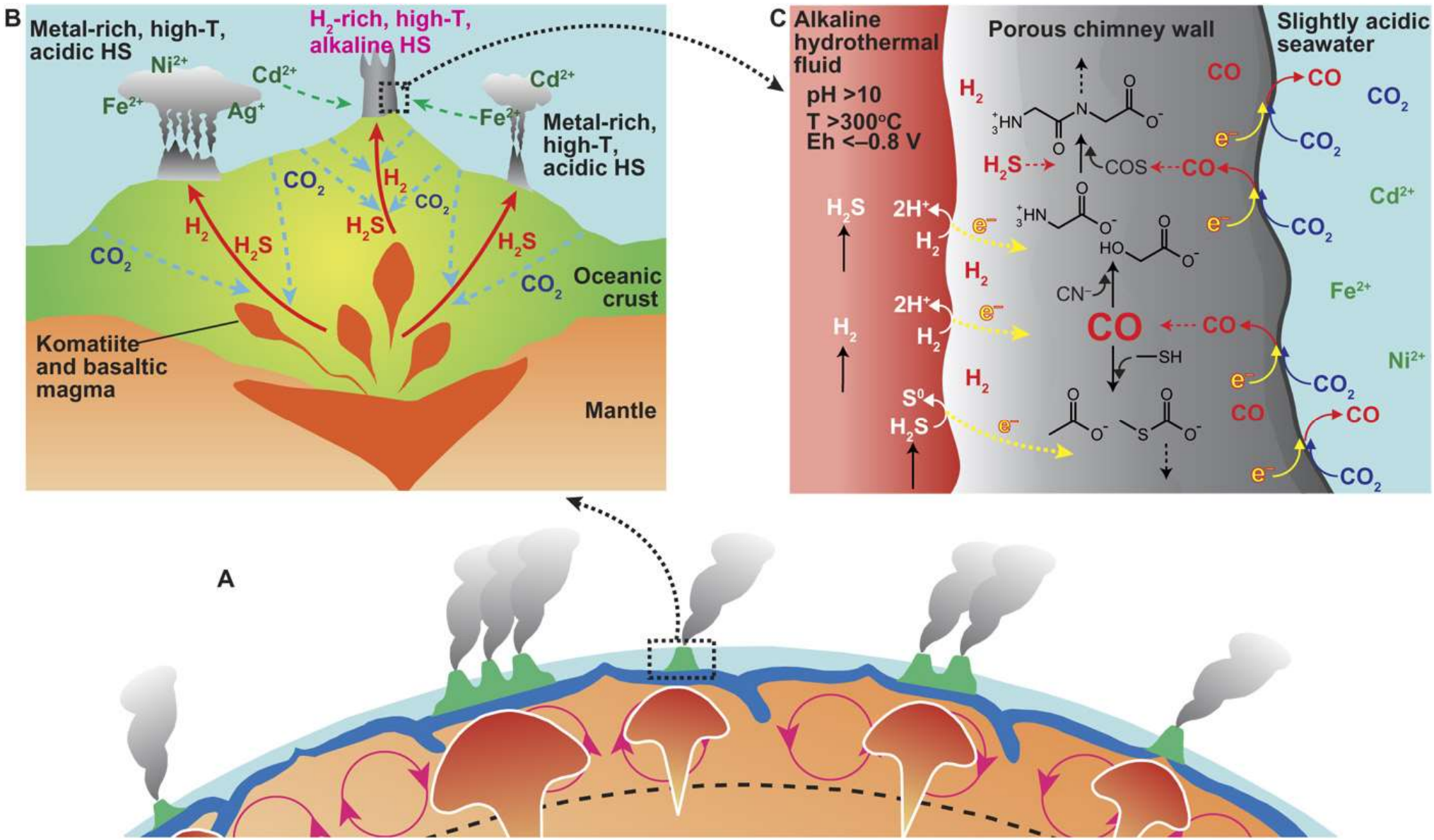
White flocculent mats in and around the extremely gassy, high-temperature (>100°C, 212°F) white smokers at Champagne Vent.



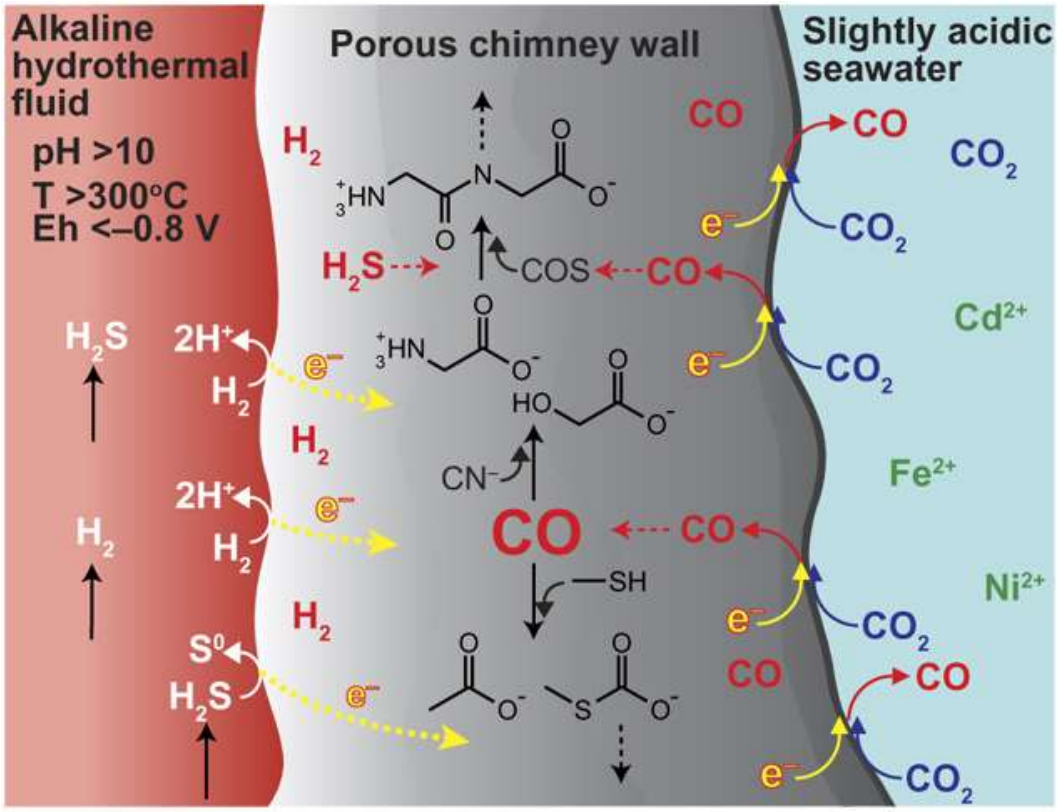
Alkaline hydrothermal vents consist of microscale caverns coated by thin membraneous metal sulfide walls → ‚Iron-sulfur world'

Deep sea vent biogeochemical cycle diagram




B
Metal-rich, high-T, acidic HS
H2-rich, high-T, alkaline HS
Ni2+
Fe2+
Ag+
Cd2+
Cd2+
Fe2+
Metal-rich, high-T, acidic HS
CO2
H2
CO2
H2
H2S
H2S
CO2
H2S
CO2
Oceanic crust
Komatiite and basaltic magma
Mantle
C
Alkaline hydrothermal fluid
pH >10
T >300°C
Eh <–0.8 V
Porous chimney wall
Slightly acidic seawater
H2
H2S
COS
CO
CO2
Cd2+
Fe2+
Ni2+
2H+
H2
CN–
SH
S0
e–
A

Deep sea vent origin of life

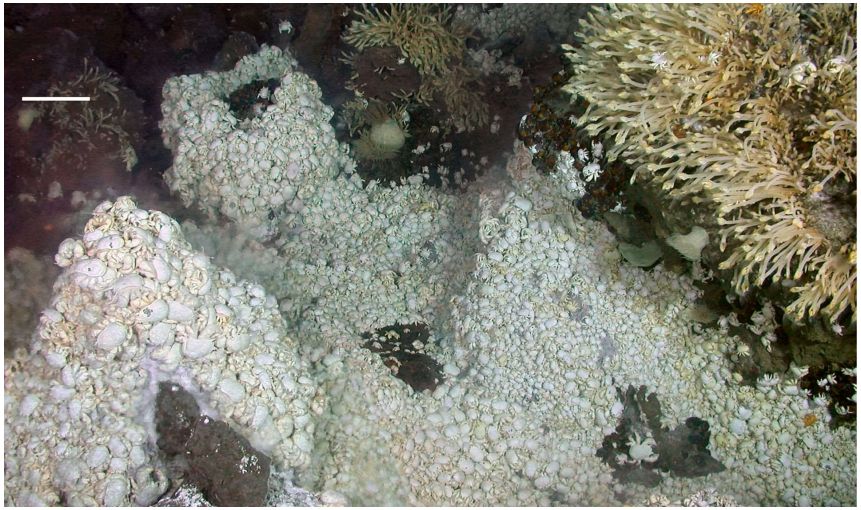


Abiotic carbon fixation in the primitive hydrothermal system.

On the ocean floor, mixing of the hydrothermal fluids and seawater generated sulfide-rich chimneys, and the potential gradient across the chimney drove a continuous electron flow. The electric potential at the chimney-seawater interface could reach less than –1 V (versus SHE) in alkaline hydrothermal vent environments. The low potential, in the presence of sulfides rich in Cd^{2+} and Ag^{+}, allowed the electrochemical CO_2 reduction to CO with the FE as high as dozens of percent, together with H_2 evolution. The produced CO served as a driving force for the subsequent abiotic organic synthesis that preceded the origin of life as schematically indicated in the figure

Kitadai et al., *Sci. Adv.* **2018**; 4: eaao7265

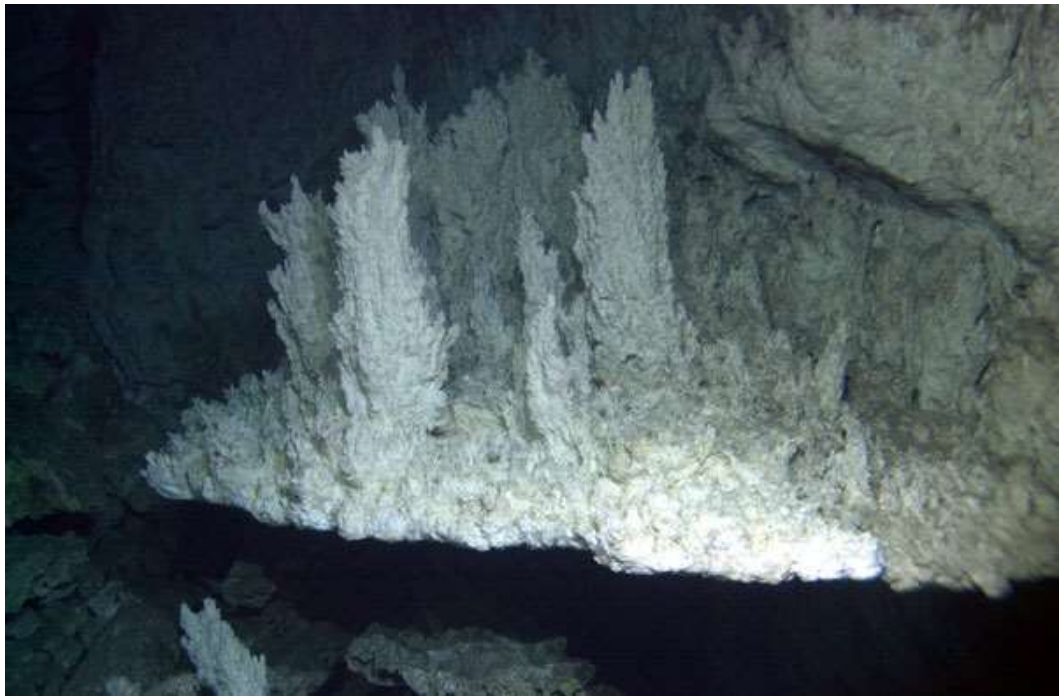
Deep sea vent fauna



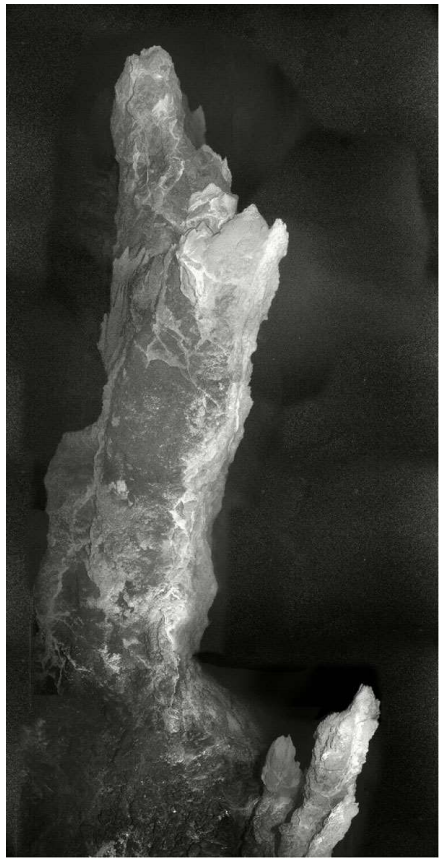
A dense fauna (*Kiwa* anomurans and *Vulcanolepas* like stalked barnacles) near East Scotia Ridge vents

Giant tube worms (*Riftia pachyptila*) cluster around vents in the Galapagos Rift

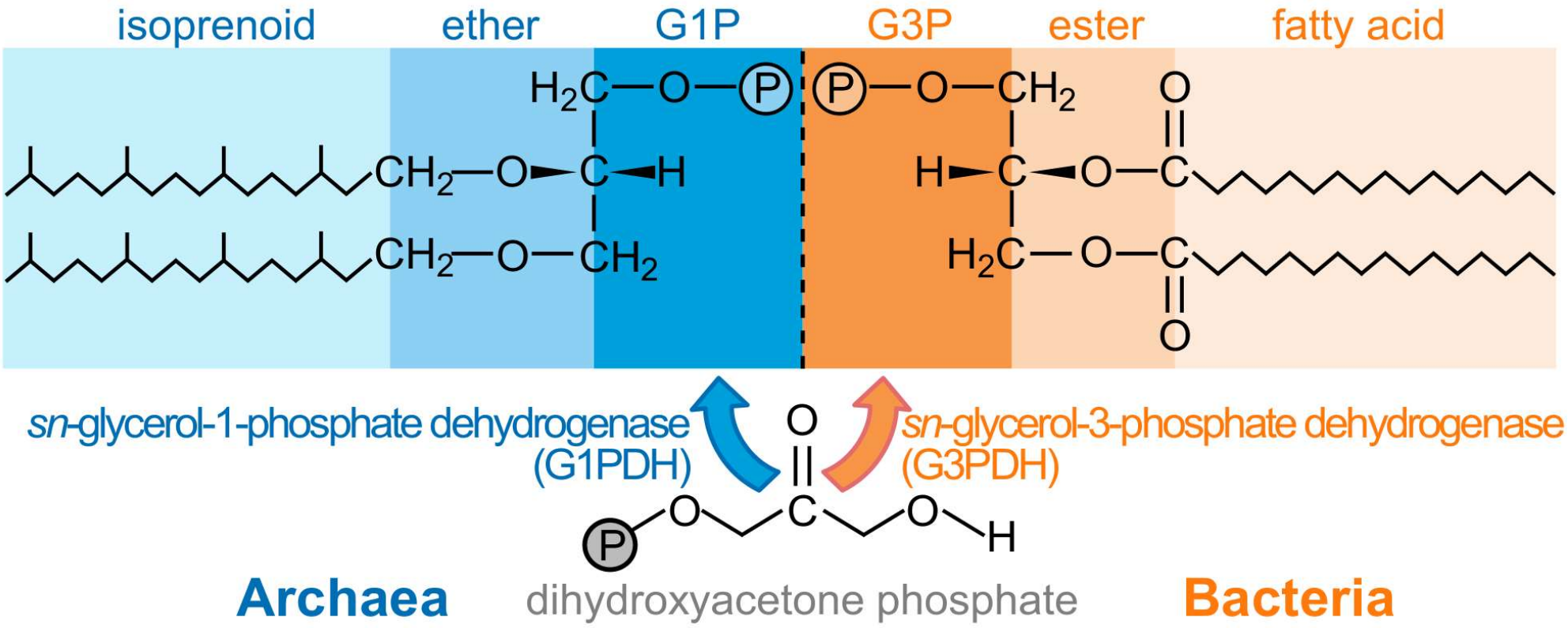
„Lost city" – white smokers: alkaline hydrothermal vents



A 1.5-meter-wide ledge on the side of a chimney is topped with dendritic carbonate growths that form when mineral-rich vent fluids seep through the flange and come into contact with the cold seawater.



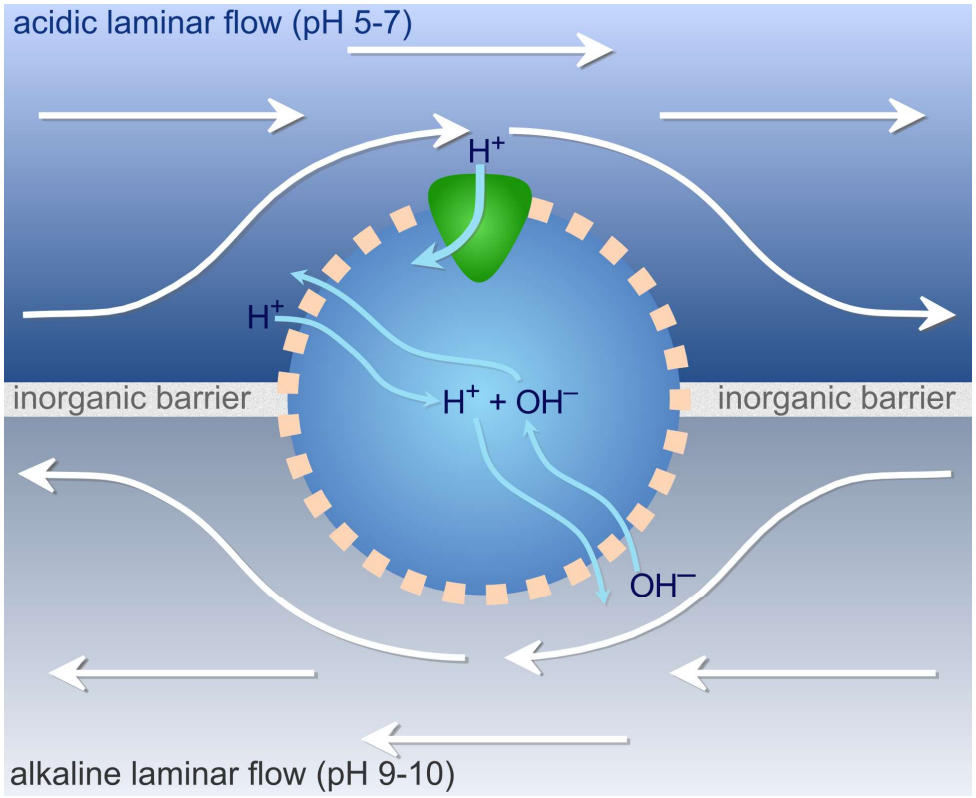
A carbonate chimney more than 9 meters (30 feet) in height. The white, sinuous spine is freshly deposited carbonate material. The top shows evidence of collapse and re-growth, as indicated by the small newly developed cone on its top



Archaeal lipids: isoprenoid chains + ether bonds + *sn*-glycerol-1-phosphate (G1P) backbone.
Bacterial lipids: fatty acids + ester linkage + *sn*-glycerol-3-phosphate (G3P) skeleton.

Despite widespread horizontal gene transfer, no bacterium has been observed with the archaeal enantiomer, or vice versa.
(ether linkages have been observed in bacterial membranes and isoprenoids are common to all three domains)

V. Sojo, A. Pomiankowski, N. Lane *PLOS Biology,* **2014**, *12(8),* e1001926

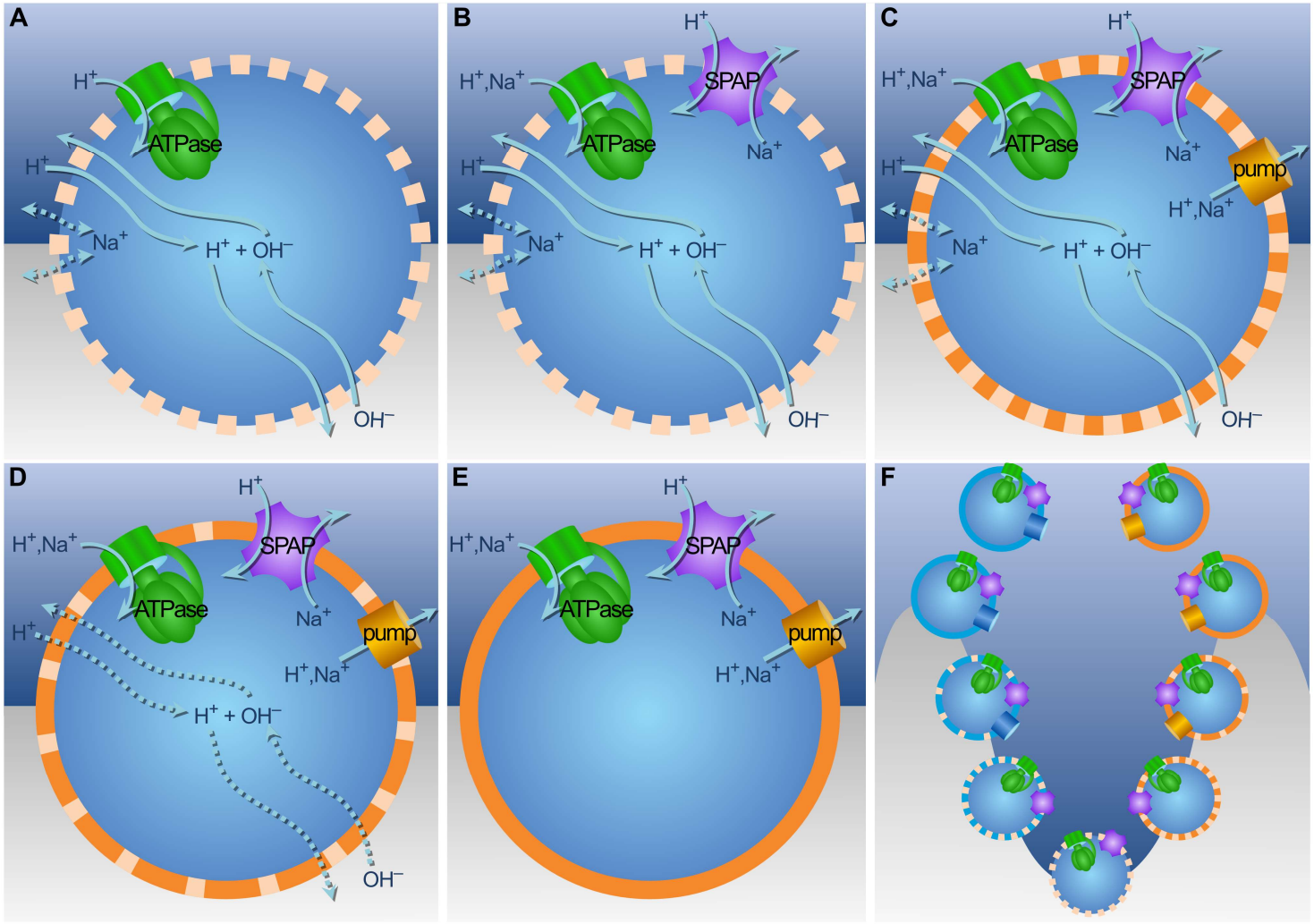


A cell with a semi-permeable membrane at the interface between an alkaline and an acidic fluid (separated elsewhere with an inorganic bareer. H^+, OH^-, Na^+, K^+, Cl^- and other ions flow according to their natural gradients.
Inside the protocell, H^+ and OH^- can neutralize into water, or leave towards either side.

A protein capable of exploiting the natural proton gradient sits on the acidic side, allowing energy assimilation via ATP production, or carbon assimilation via CO_2 fixation.

V. Sojo, A. Pomiankowski, N. Lane *PLOS Biology,* **2014**, *12(8),* e1001926

The role of sodium-proton antiporter (SPAP)



A) H^+ gradient drives energy metabolism (ATPase) or carbon metabolism (Ech)

B) SPAP generates Na^+ from H^+ gradient

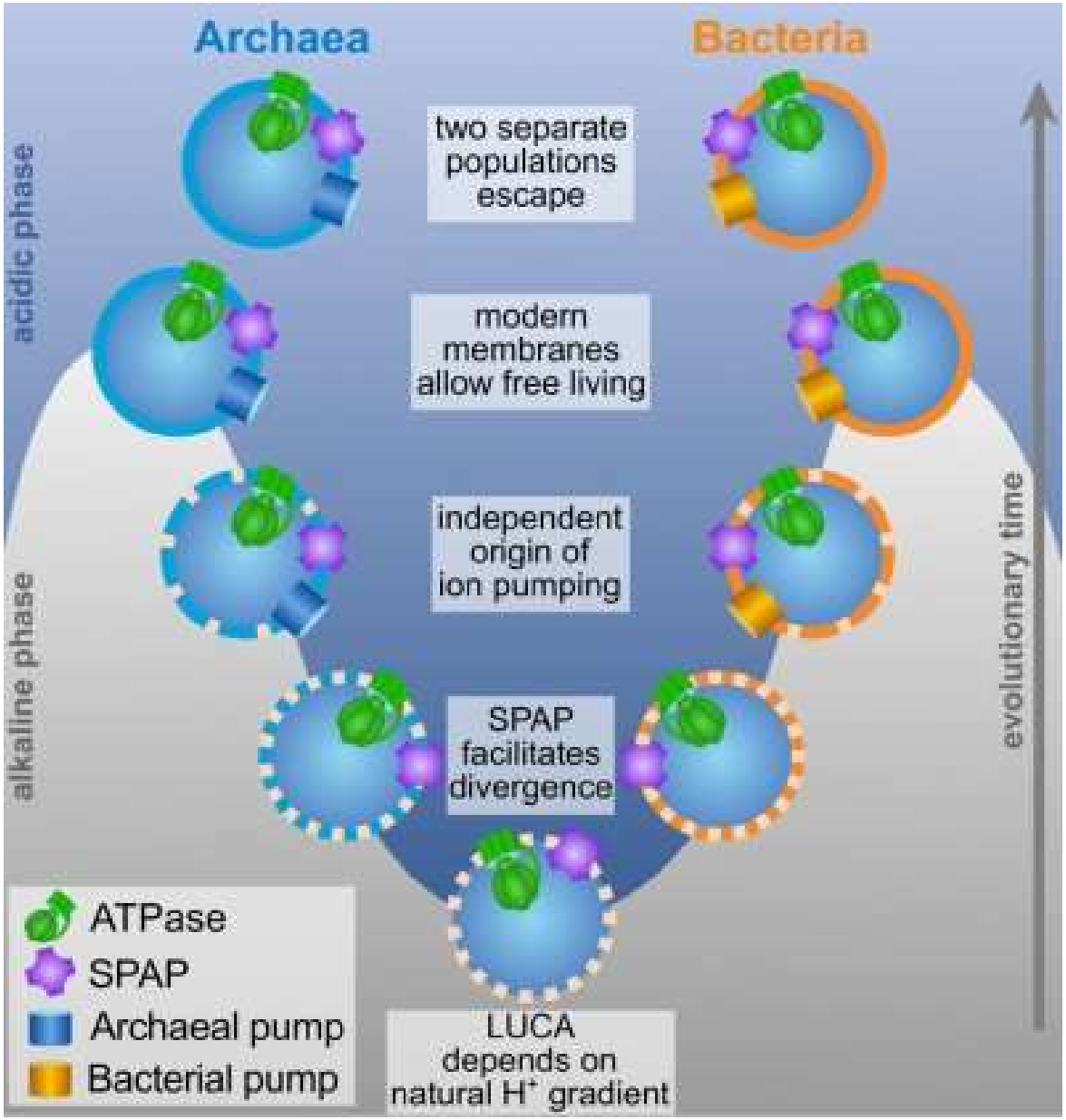
C) Membrane pumps secret H^+ and Na^+

D) Tighter membranes are now produced, to colonize less alkaline environments

E) Impermeable membranes → gradients created by proteins, independently from the natural environmental gradients

F) SPAP favors divergence, selection for active pumping and tighter membranes; independent evolution of archea and bacteria

V. Sojo, A. Pomiankowski, N. Lane
PLOS Biology, **2014**, *12(8),* e1001926



Origin of autotrophy and development of cell membrane

Ion pumping and phospholipid membranes evolved independently in bacteria and archea.

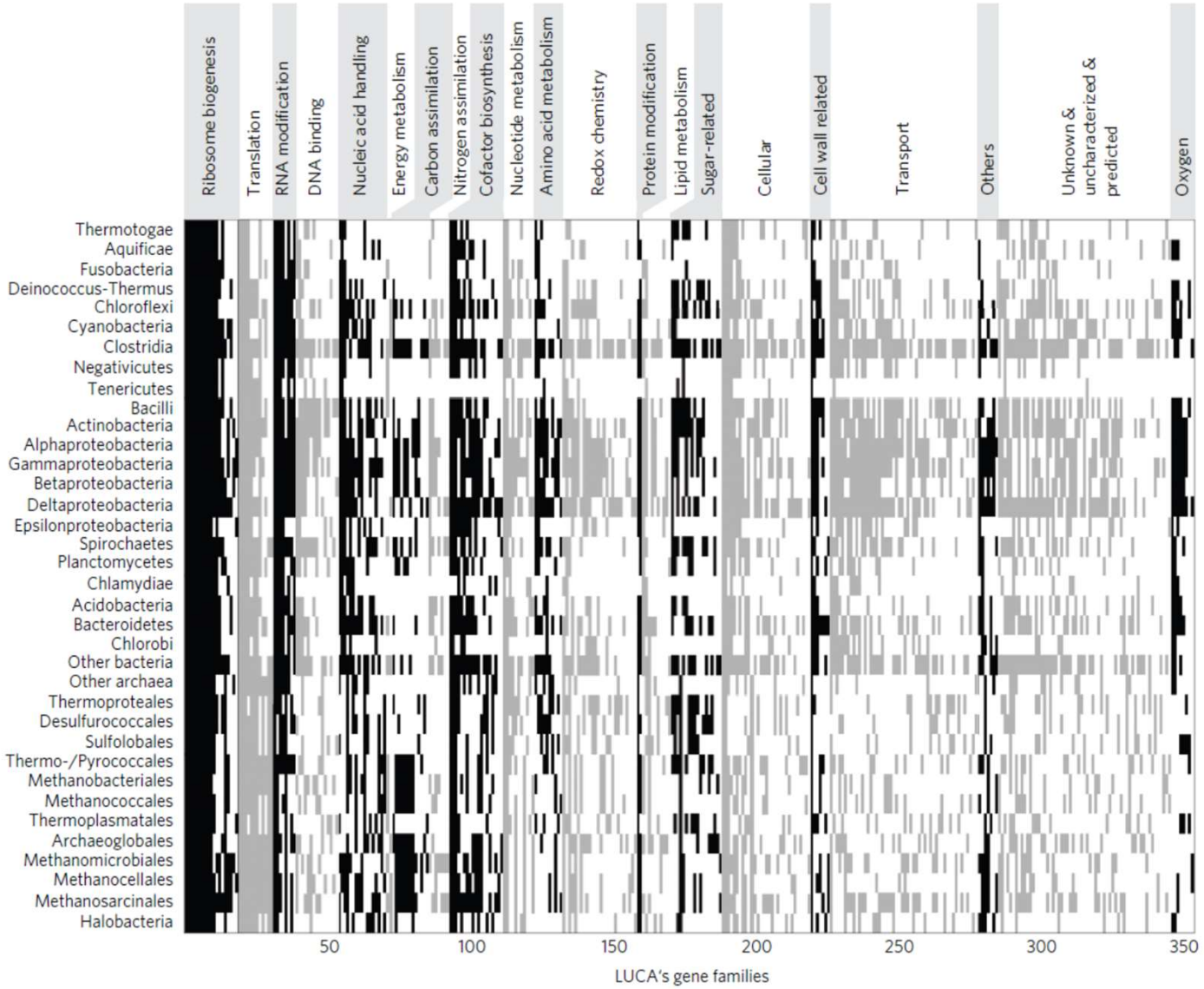
Energy could have been delivered by the natural proton gradient in alkaline hydrothermal vents, if the membrane was much more leaky than contemporary ones.

Development of proton pumping allowed for escape from the vent environment.

sodium-proton antiporter (SPAP)

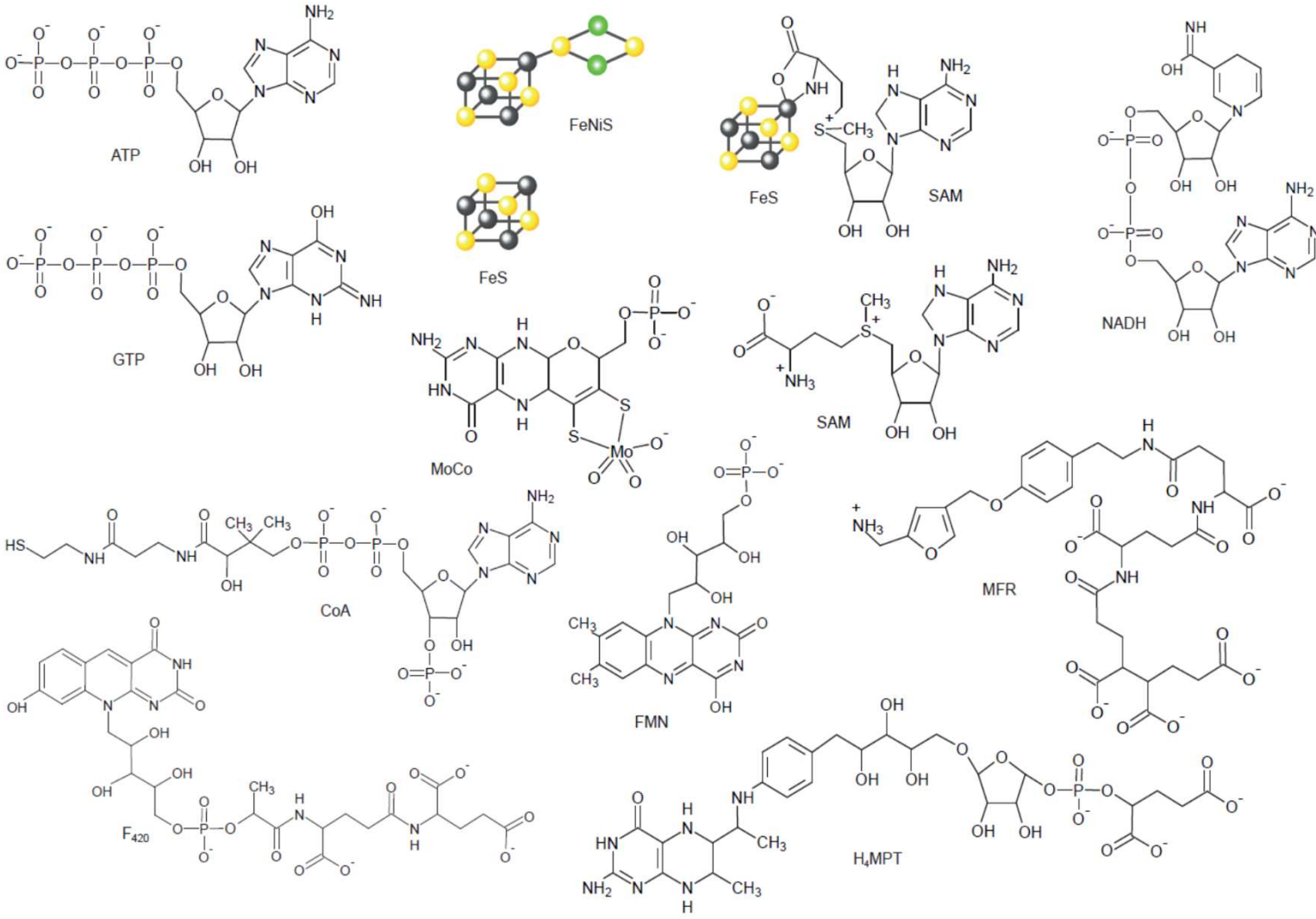
V. Sojo, A. Pomiankowski, N. Lane
PLOS Biology, **2014**, *12(8),* e1001926

Taxonomic distribution of LUCA's genes grouped by functional categories



M.C. Weiss et al. *Nature Microbiology,* **2016**, *Article 16116*

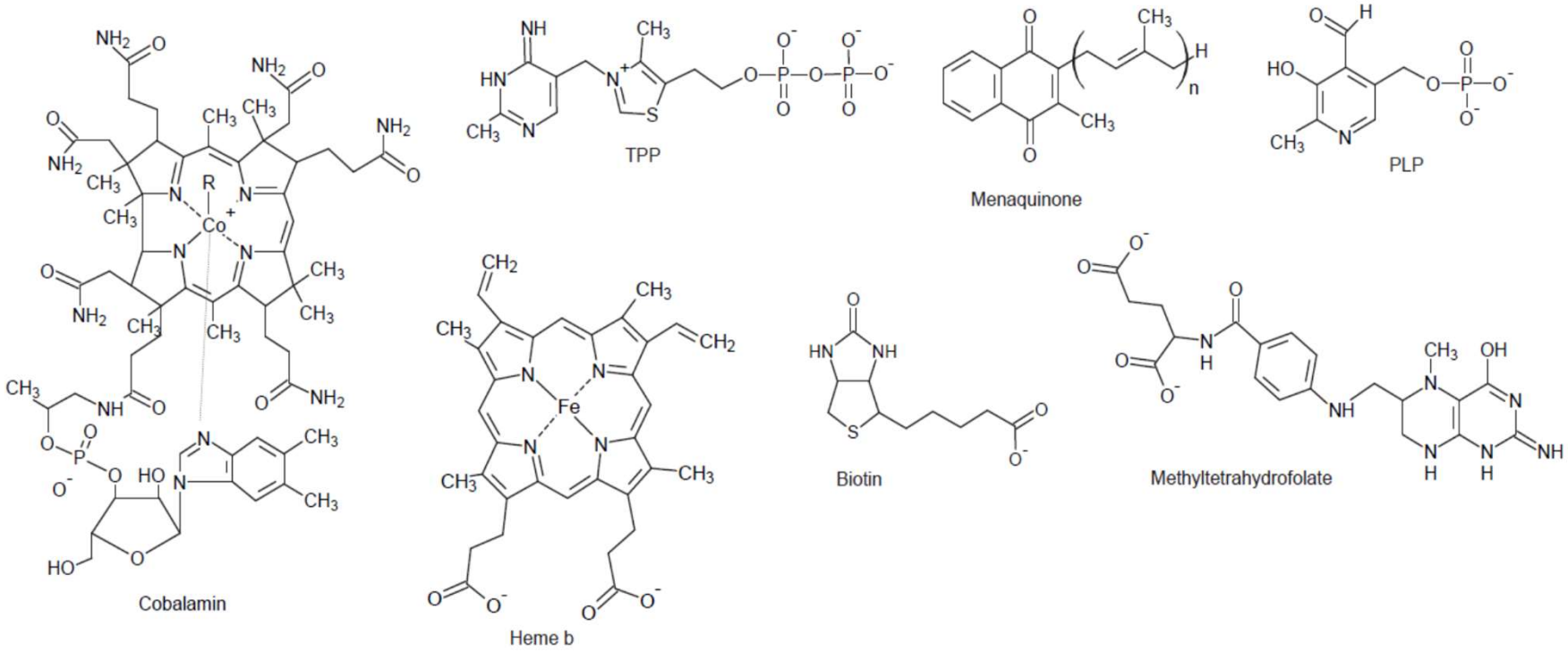
Structures of the cofactors found in LUCA's protein set.



FeNiS – nickel-iron-sulfur cluster
FeS – iron-sulfur cluster
MoCo – molybdenum cofactor
SAM – S-adenosylmethionine
CoA – coenzyme A
MFR – methanofuran
H4MPT – tetrahydromethanopterin
TPP - thiamine pyrophosphate
PLP - pyridoxal phosphate
NTP – nucleoside triphosphate.

M.C. Weiss et al. *Nature Microbiology,* **2016**, *Article 16116*

Structures of the cofactors found in LUCA's protein set.



Mononuclear metal centers (Fe and Cu) and the non-standard amino acid selenocysteine are not shown, nor are small protein electron carriers such as ferredoxin or rubredoxin. NTP is also listed as a cofactor, but not shown here as it stands for any of the nucleoside triphosphates in those cases when it's not known which one is bound by the enzyme, or when more than one nucleoside triphosphate can be used

M.C. Weiss et al. *Nature Microbiology,* **2016**, *Article 16116*

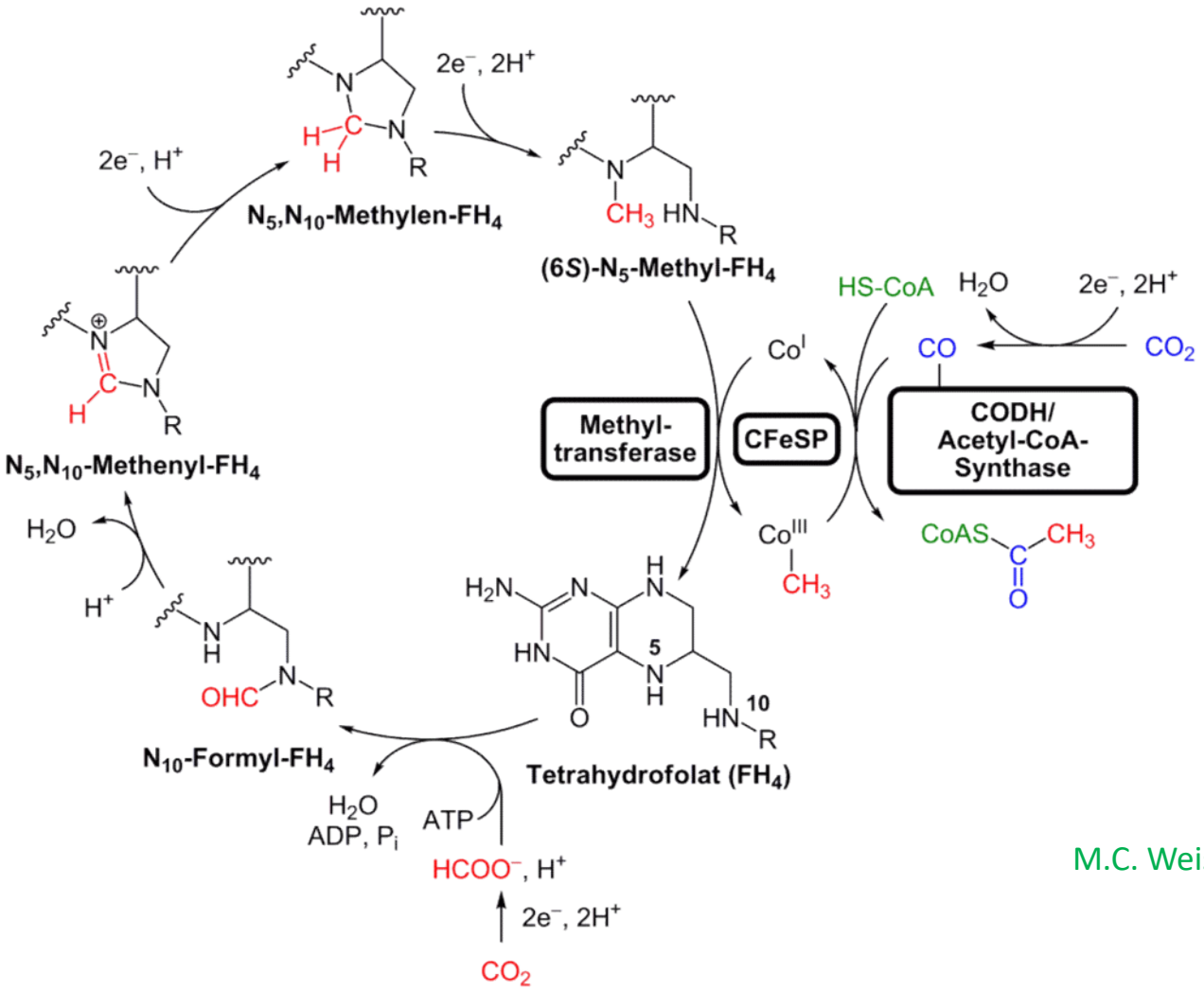
Phylogenetic identification of LUCA's proteome

355 protein families shared among contemporary *archaea* and *bacteria*, including:

- 19 proteins involved in ribosome biogenesis
- 8 aminoacyl tRNA synthethases
- proteins for carbon, energy, and nitrogen metabolism
- rotor-stator ATP synthase subunit (ion gradients were likely supplied geochemically)
- substrate-level phosphorylation (acetylphosphate from acetyl-CoA)
- reverse gyrase – specific for currently living hyperthermophilic organisms
- chemolitoautotrophy enzymes present (WL pathway), chemoorganoautotrophy enzymes absent

M.C. Weiss et al. *Nature Microbiology,* **2016**, *Article 16116*

Wood-Ljungdahl (WL) anaerobic pathway of carbon fixation



M.C. Weiss et al. *Nature Microbiology,* **2016**, *Article 16116*

A primitive metabolic pathway for carbon fixation, still used by some contemporary chemoautotrophic organisms

Metabolism of LUCA

Among six currently known pathways of CO_2 fixation, only WL pathway was present in LUCA:

The relevant enzymes are packed with FeS and FeNiS centres

They require cofactors: flavin, F_{420}, methanofuran, two pterins and corrins

Hydrogenases also present in LUCA's genome → electrons likely obtained from hydrogen, as in modern microbes using the WL pathway

Nitrogenase and glutamine synthethase serve for nitrogen fixation

WL pathway, nitrogenase and hydrogenases are very oxygen-sensitive
→ LUCA was an anaerobic autotroph that could live from gases H_2, CO_2, and N_2.

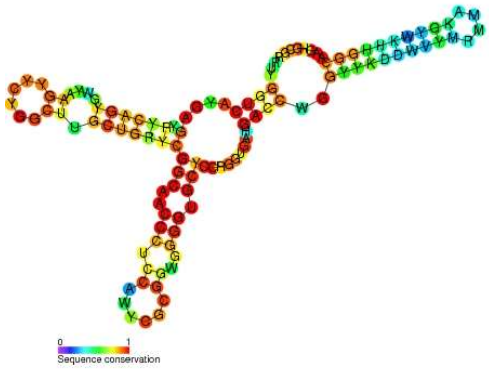
M.C. Weiss et al. *Nature Microbiology,* **2016**, *Article 16116*

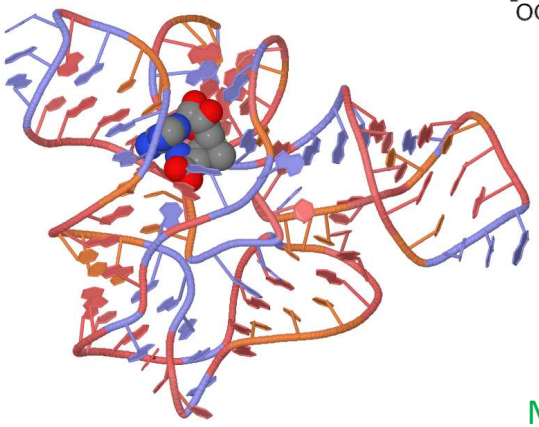
Metabolism of LUCA

Enzymes for cofactor biosynthesis, including pterins, MoCo, cobalamin, siroheme, TPP, CoM and F_{420}, are also conserved.

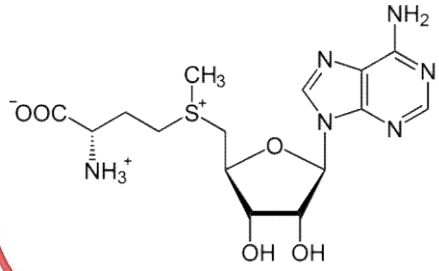
Many of them are S-adenosyl methionine(SAM)-dependent

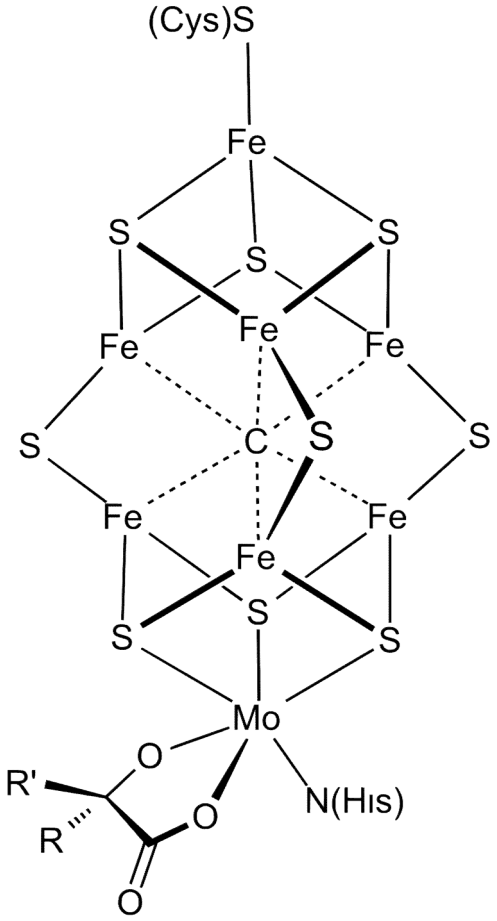
SAM chemistry is based on oxygen-sensitive FeS-containing proteins that initiate radical-dependent methylations.





SAM-binding riboswitches



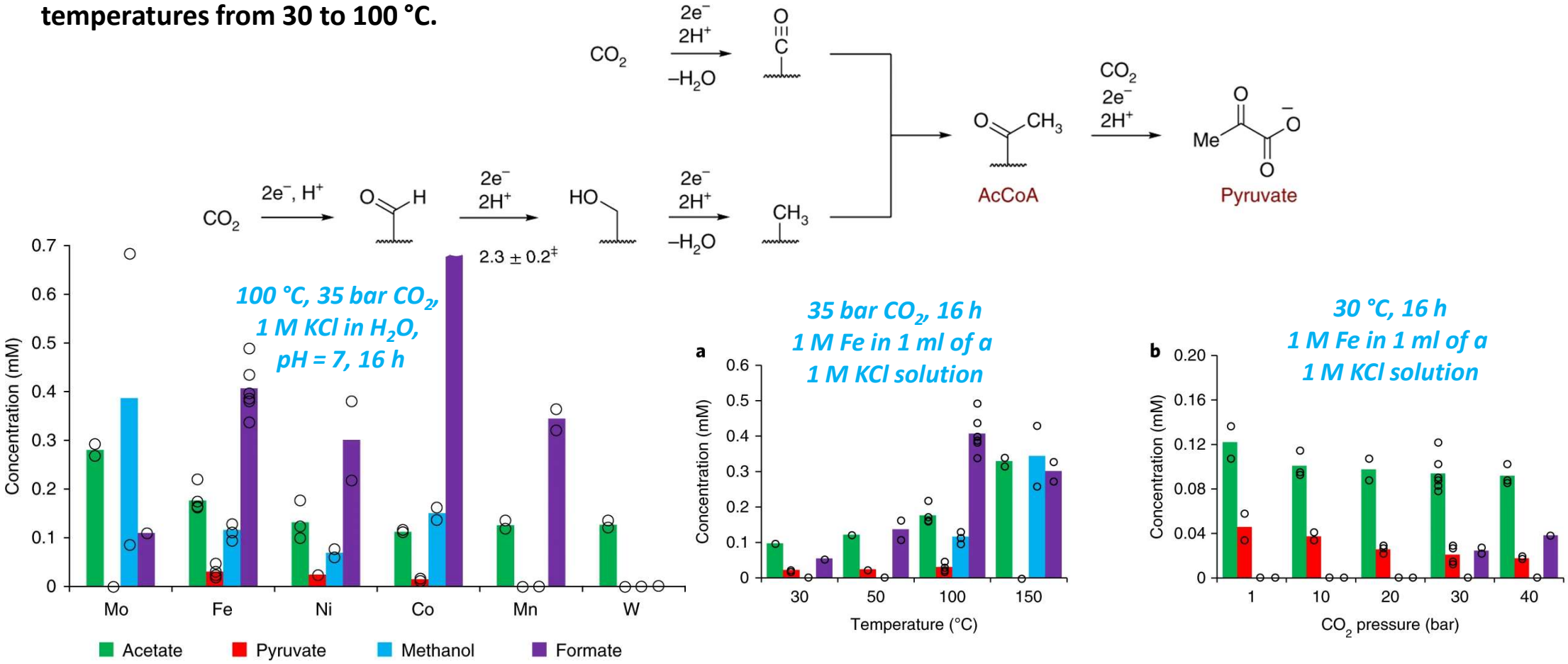


FeMo cofactor of nitrogenase

M.C. Weiss et al. *Nature Microbiology,* **2016**, *Article 16116*

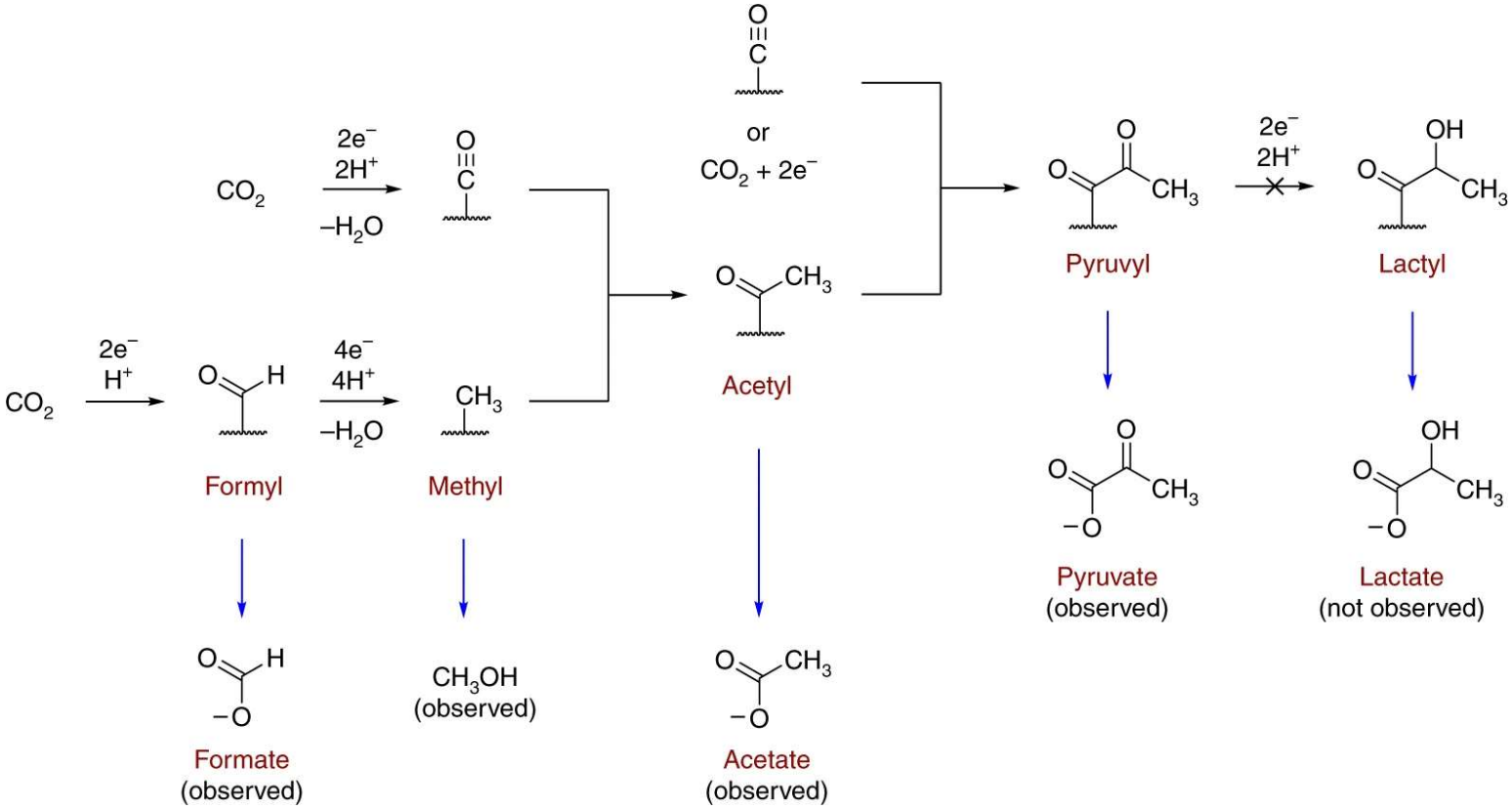
Prebiotic Wood-Ljungdahl (WL) pathway analogue

native transition metals (Fe^0, Ni^0 and Co^0) selectively reduce CO_2 to acetate and pyruvate—the intermediates and end-products of the AcCoA pathway—in near millimolar concentrations in water over hours to days using 1–40 bar CO_2 and at temperatures from 30 to 100 °C.



S.I. Varma, K.B. Muchowska, P. Chatelain, J. Moran *Nature Ecology and Evolution,* **2019**, *2,* 1019-1024

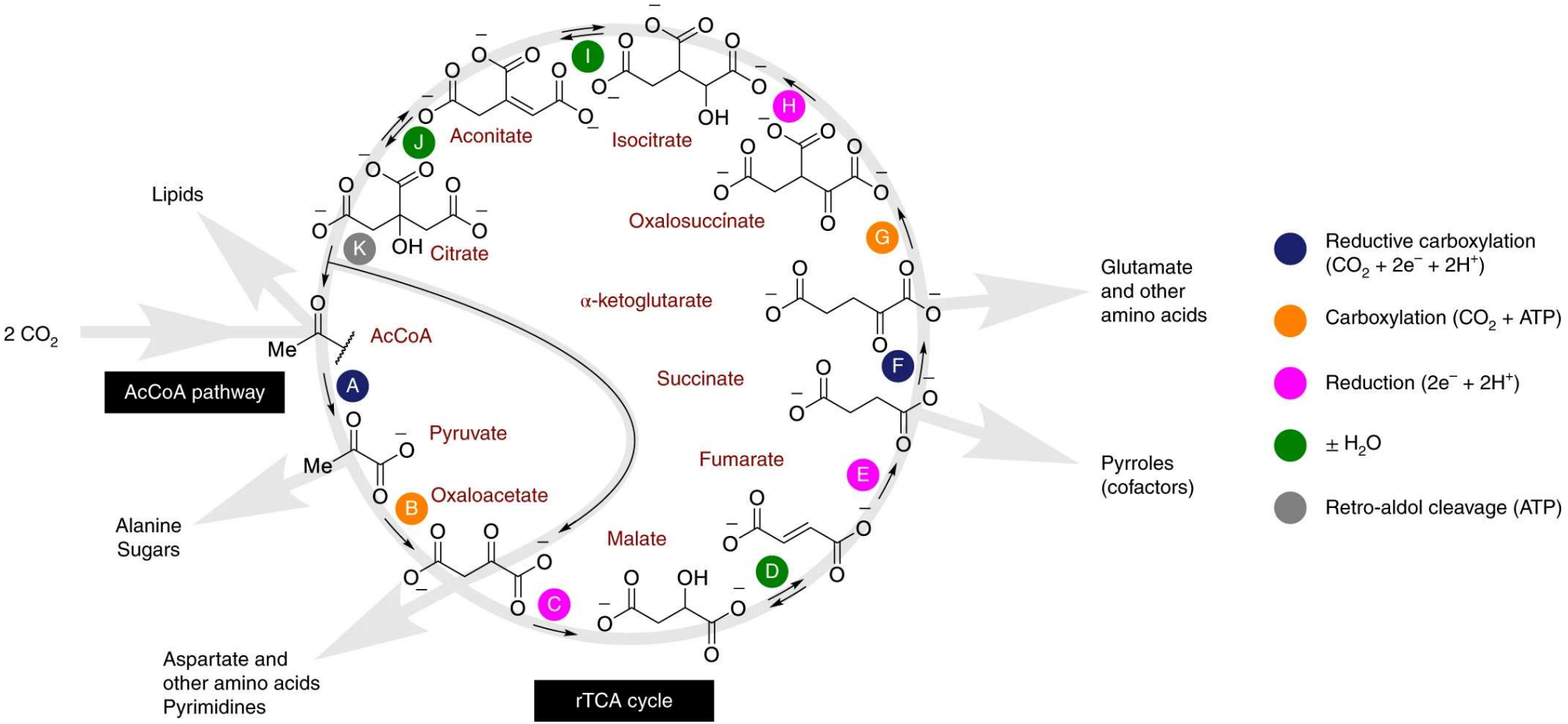
Prebiotic Wood-Ljungdahl (WL) pathway analogue



Plausible mechanism for carbon fixation on the surface of Fe^0 accounting for the detection of formate, methanol, acetate and pyruvate in aqueous solution upon hydrolysis with KOH

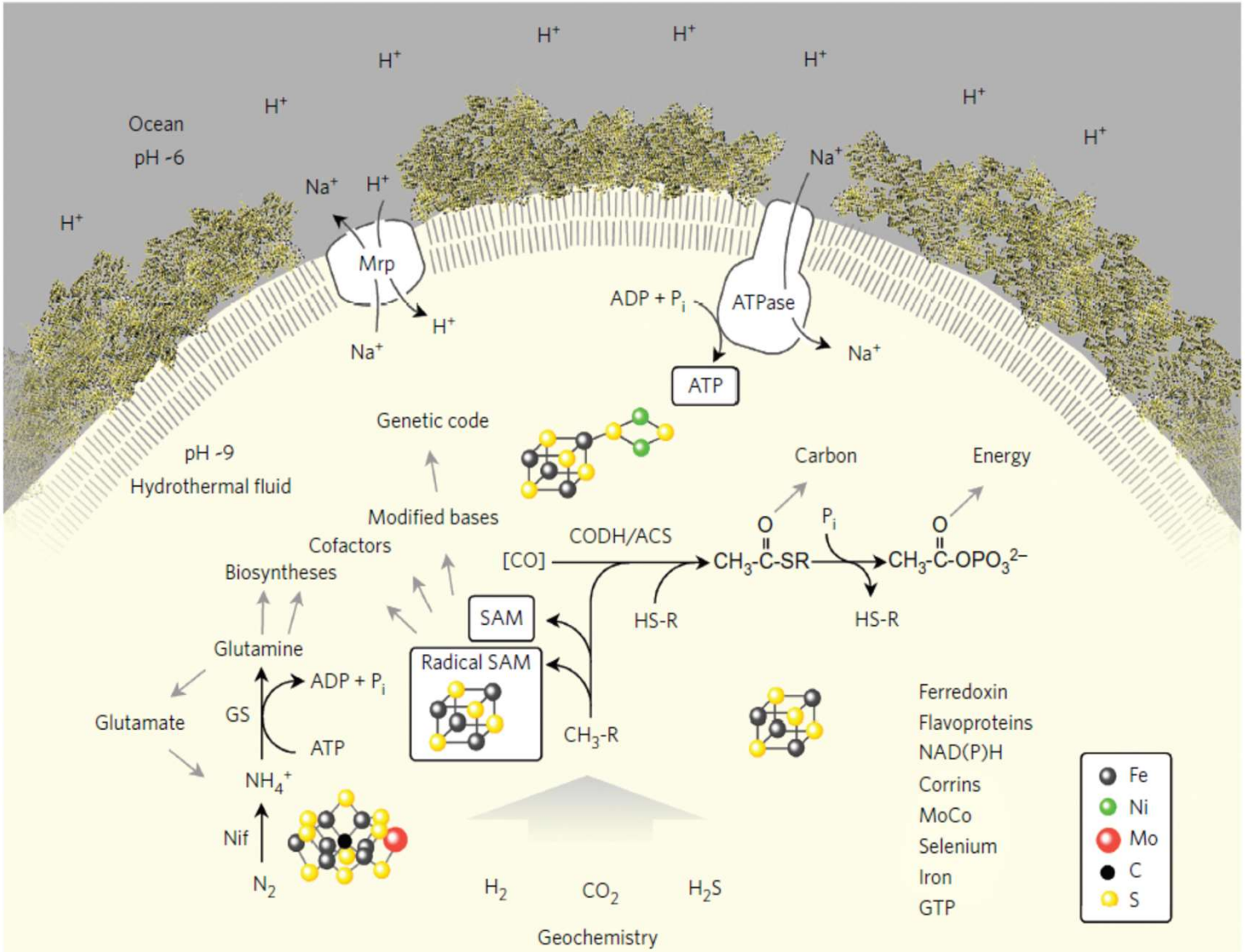
S.I. Varma, K.B. Muchowska, P. Chatelain, J. Moran *Nature Ecology and Evolution,* **2019**, *2,* 1019-1024

Prebiotic Wood-Ljungdahl (WL) pathway analogue



Hypothetical ancestral proto-anabolic network consisting of a hybrid of the AcCoA pathway and the rTCA cycle, showing the role of its intermediates as universal biosynthetic precursors

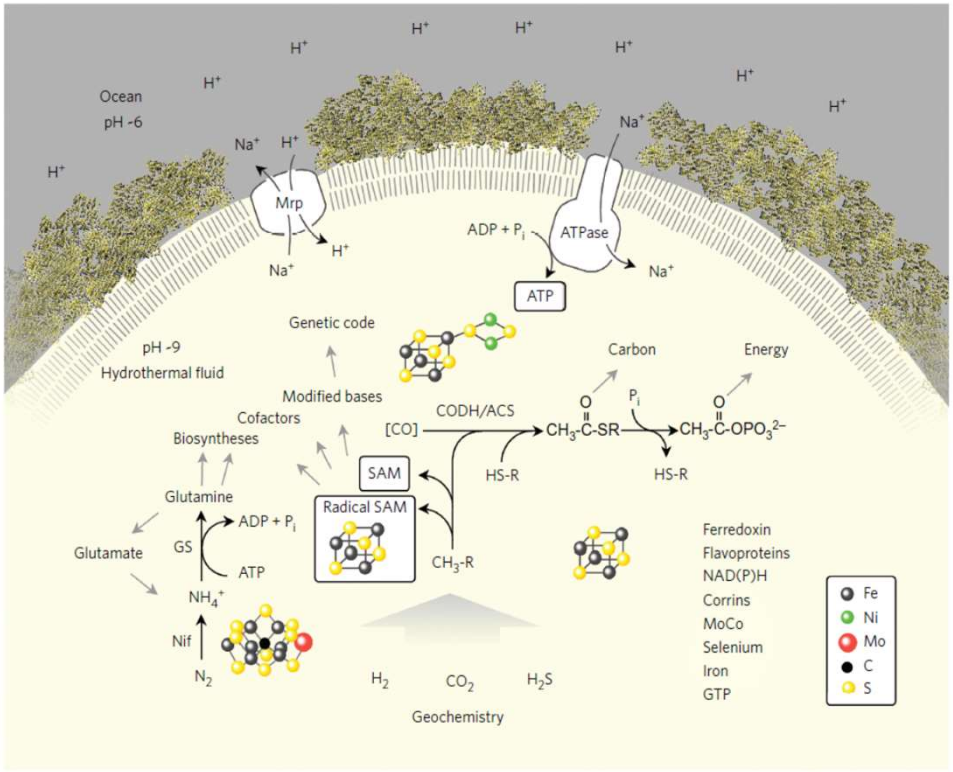
S.I. Varma, K.B. Muchowska, P. Chatelain, J. Moran *Nature Ecology and Evolution,* **2019**, *2,* 1019-1024



M.C. Weiss et al. *Nature Microbiology,* **2016**, *Article 16116*

LUCA reconstructed from the genome data

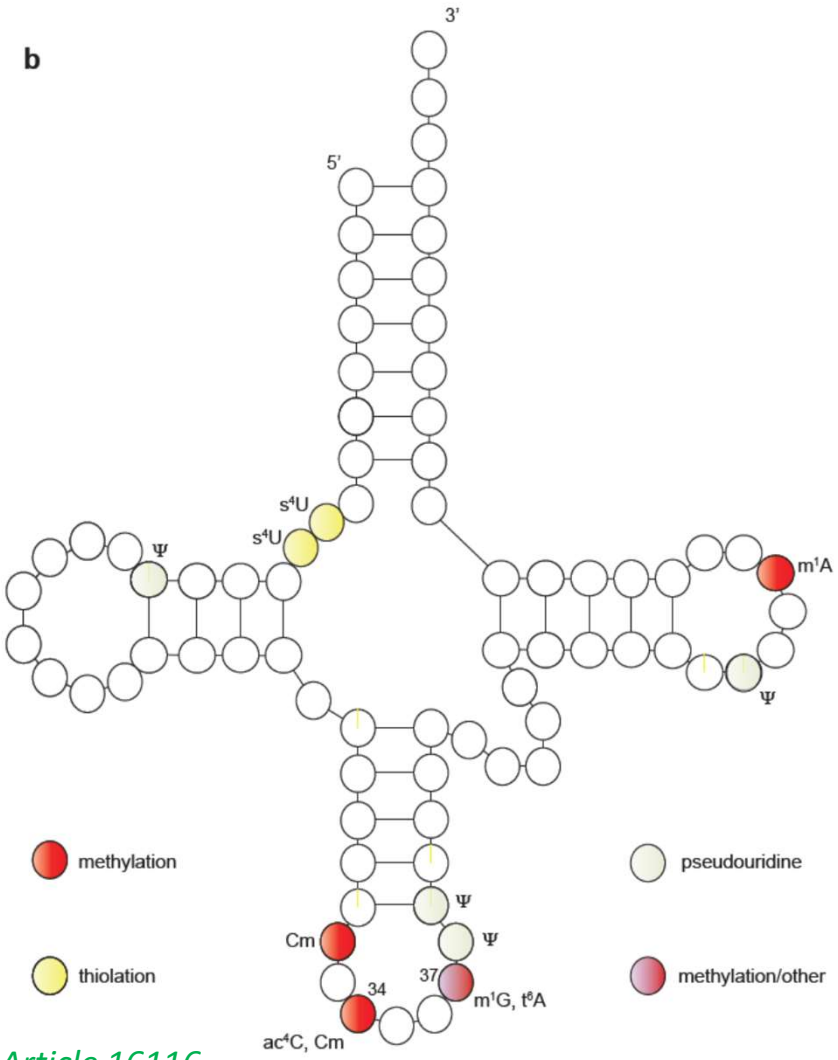
Summary of the main interactions of LUCA with its environment, a vent-like geochemical setting as inferred from genome data.



In modern CODH/ACS complexes, CO is generated from CO_2 and reduced ferredoxin.
A Na^+/H^+ antiporter could transduce a geochemical pH gradient (indicated on the left) inherent in alkaline hydrothermal vents into a more stable Na^+ gradient to feed a primordial Na-dependent ATP synthase.

Modified nucleosides and the genetic code

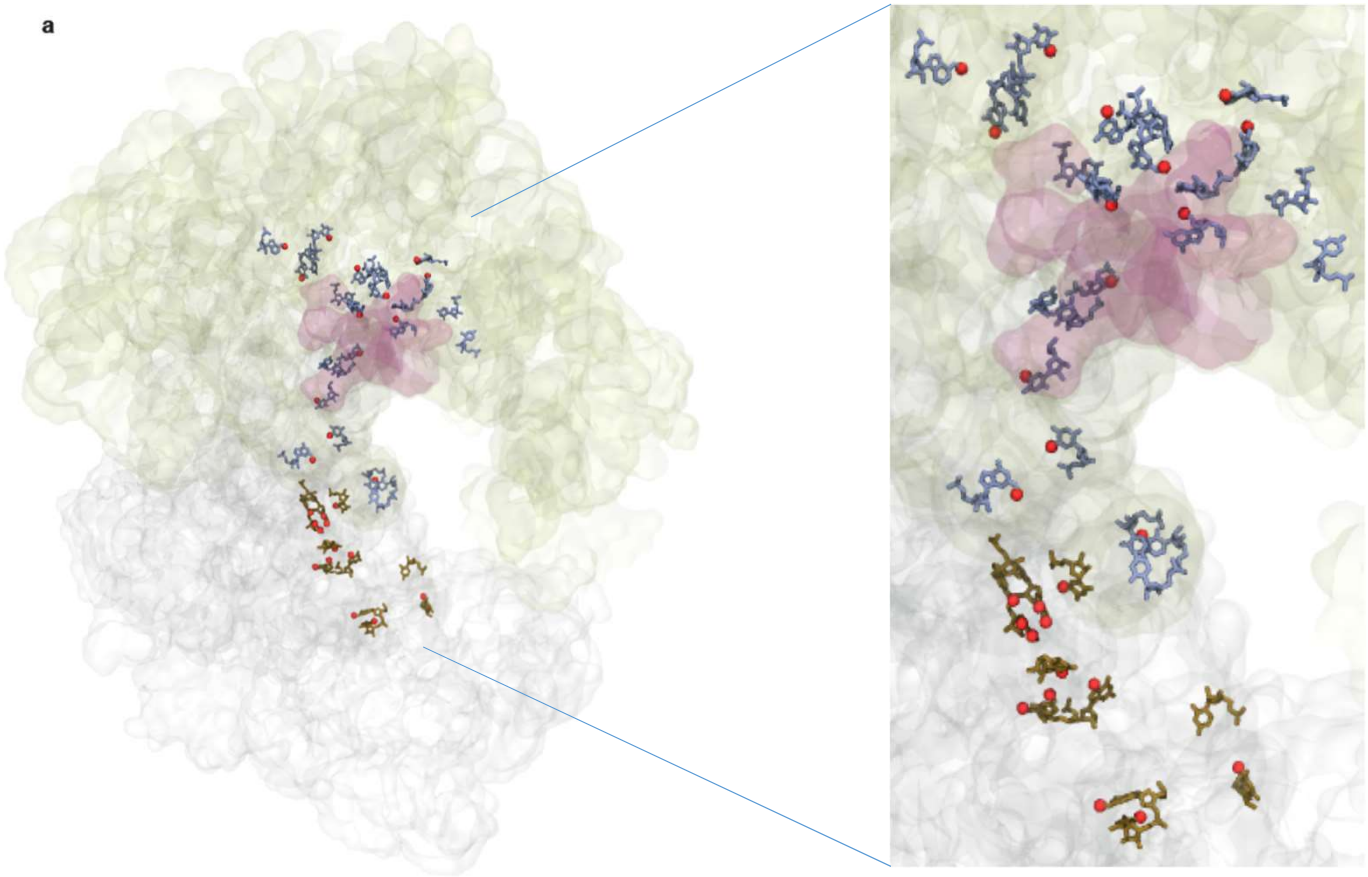
LUCA had also genes involved in RNA nucleoside modifications (mainly methylations and thiomethylations) still required today e.g. for the anticodon recognition process.



Cloverleaf secondary structure representation of tRNA showing only those posttranscriptional nucleoside modifications that are conserved among bacteria and archaea in both identity and position. (5-methoxyuridine at position 34 in archaea has been disputed).

M.C. Weiss et al. *Nature Microbiology,* **2016**, *Article 16116*

Modified nucleosides and the genetic code

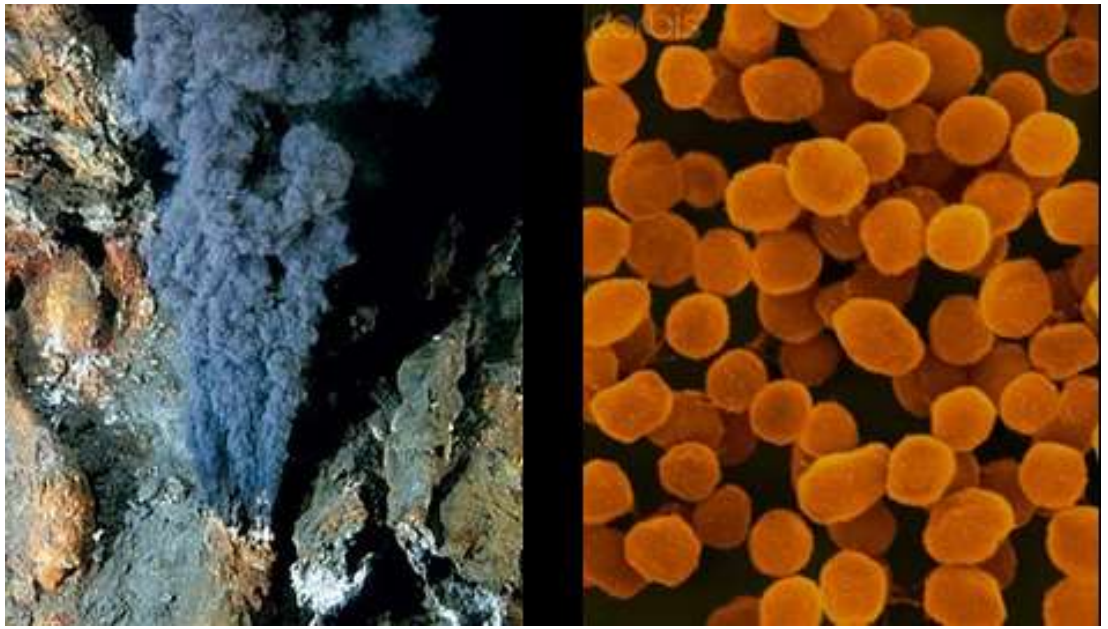


M.C. Weiss et al. *Nature Microbiology,* **2016**, *Article 16116*

Structure of the E. coli ribosome (PDB ID: 4YBB), with the large and small subunits shown in green and silver, respectively. The peptidyl-transferase site is shaded pink. The modified nucleosides of 23S rRNA are depicted in icy blue, while in 16S rRNA they are ochre. Modification of C2501 to 5-hydroxycytidine is not present in the structure. Methyl group carbons are shown as red balls.

Closest living relatives of LUCA

clostridia
anaerobic bacteria
(botulin, gangrene, tetanus)

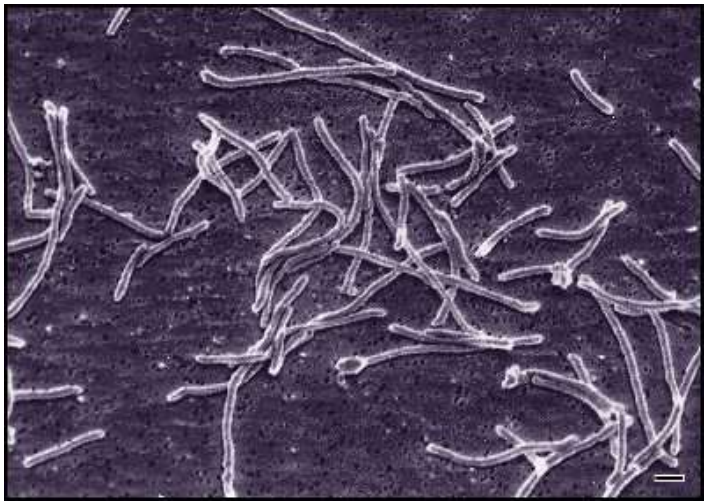


Deep ocean vent - home to the extremophilic archeon
Methanococcus jannaschii

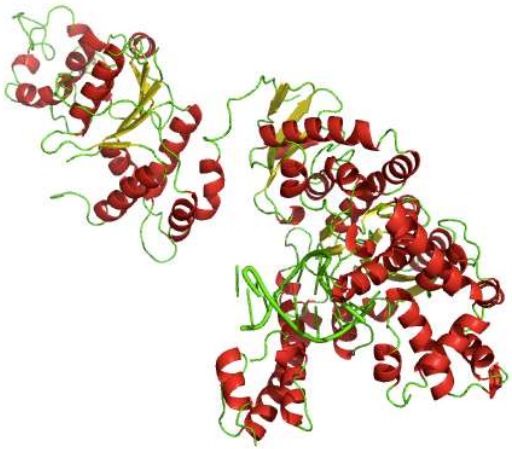
They use the WL pathway, abundant also today, some species can live from methyl groups (methane gas on marshes andwetlands), and they depend on H_2 (from geology or H_2-producing fermentation)

Geological source of hydrogen: serpentinization (iron + hot water, anoxic) $Fe^{2+} + H_2O \rightarrow Fe_3O_4 + H_2$

Thermophiles



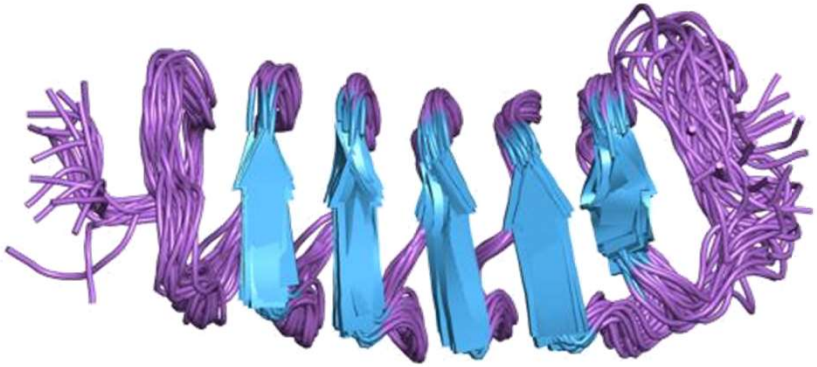
Thermus aquaticus



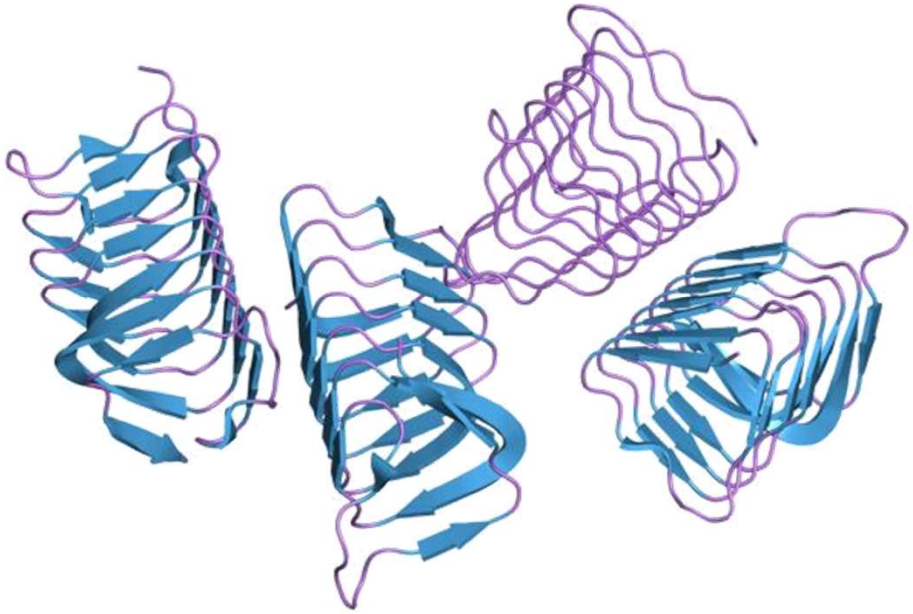
3D structure of Taq Polymerase.

Hot springs with algae and bacteria in Yellowstone National Park

Cold adaptation

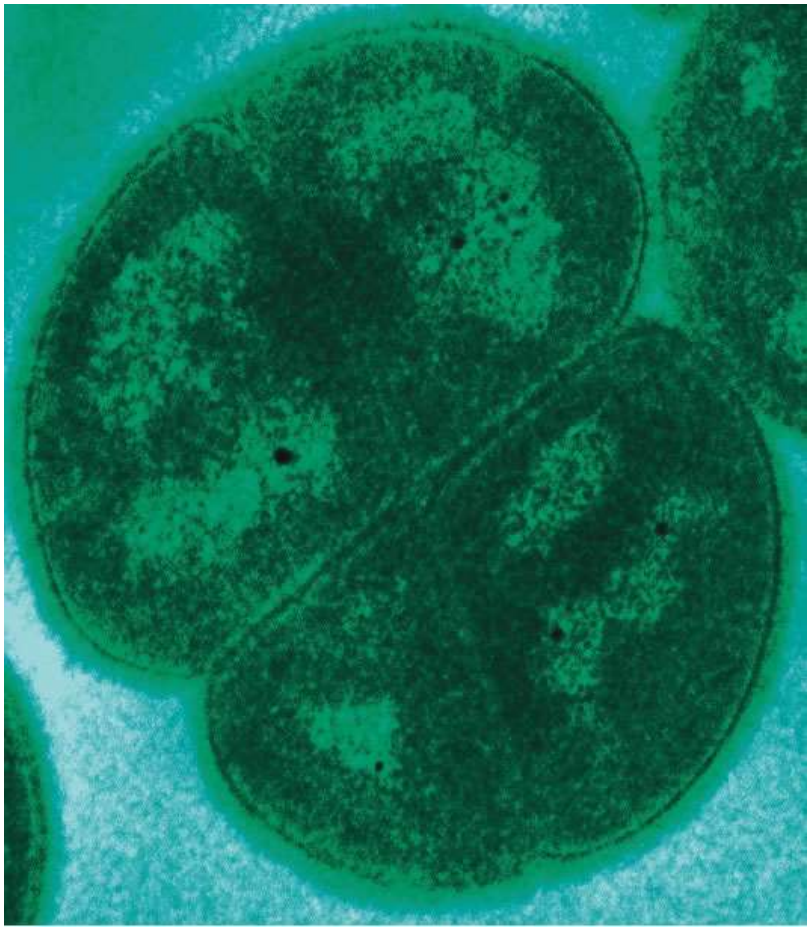


Structure of the *Tenebrio molitor* beta-helical antifreeze protein



Structure of *Choristoneura fumiferana* (spruce budworm) beta-helical antifreeze protein

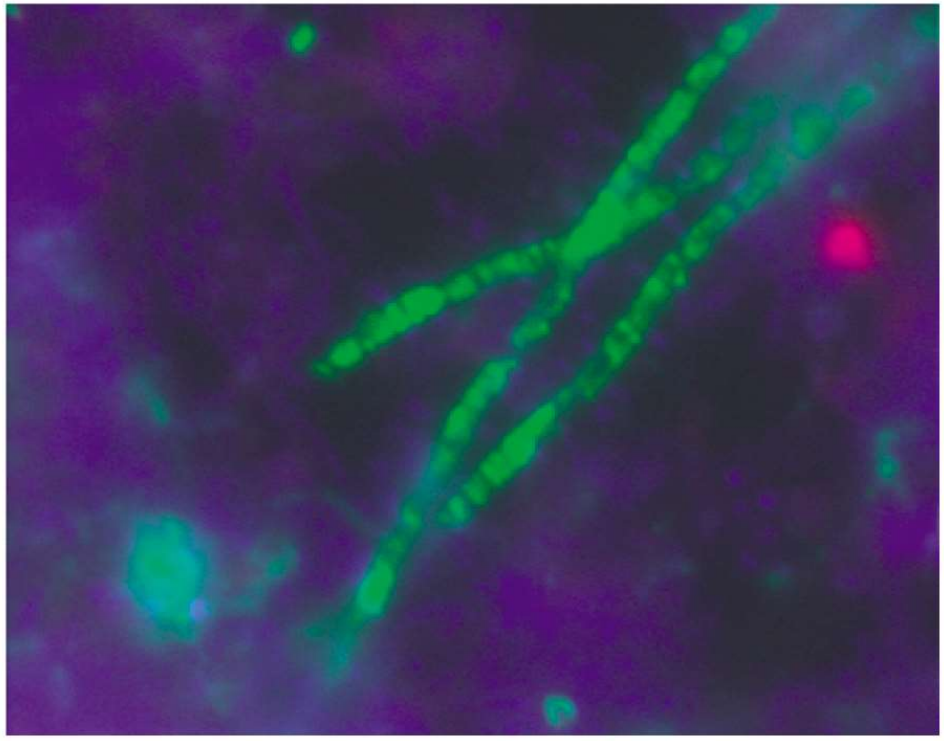
Drought, salinity, radiation



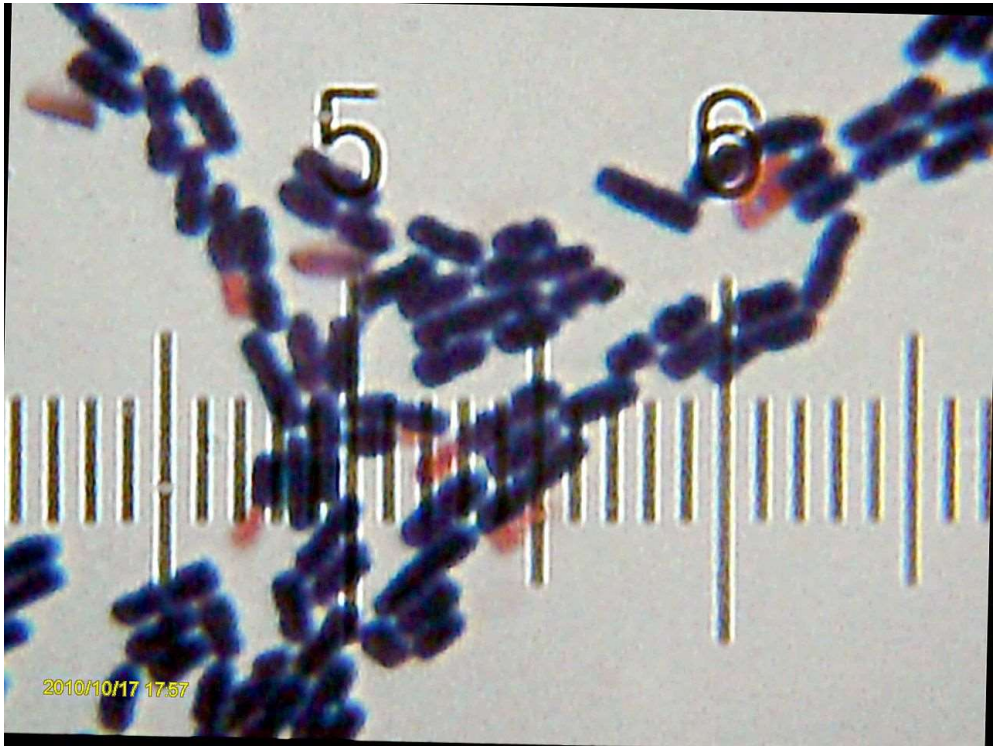
A tetrad of D. *radiodurans*

Efficient DNA damage repair,
Trehalose as the main sugar – glass solid, no crystallization

Acid, base



Acidobacterium



A typical *bacillus* culture. Many alkaliphiles possess a *bacillus* morphology