

Paradoxes of molecular biosignatures

Molecular systems that cannot possibly arise *without* life

The paradox: Such a life form could never arise.

For example, amino acids are bad biosignatures, as they can arise abiologically

Even enantiomerically enriched amino acids seem to arise without life (Sandra Pizzarello)

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The paradox is naively formulated

The (in)organic molecules are formed universally

We then look for Darwinian capability within them

A biopolymer requires homochiral building blocks to be evolvable. Further, it must be build from a controlled library.

What does Darwinism need by way of a genetic molecule?

- (a) A one-dimensional biopolymer*, that...
- (b) can change its structure to change its information, but ...
- (c) any changes in the structure of the biopolymer *cannot* change its physical chemical behavior substantially, e.g.: *its solubility, its molecular recognition, reactivity*

* We have tried to assemble a two dimensional Cairns-Smith life form; so far no luck.

Data show that such systems are scarce ...

Proteins, polysaccharides, most every other class of molecules, including abiological polymers, physical behavior and reactivity change dramatically even with small sequence changes.



Sickle cell hemoglobin. Change one lousy amino acid in 576, and the protein precipitates.



If DNA/RNA precipitated every time a nucleotide changed, evolution would grind to a halt

A genetic biopolymer must have fairly constant properties if it is to support Darwinism, where it is replicated with imperfections, with the imperfections themselves replicable

Pretty much true with DNA and RNA

Pretty much every DNA/RNA sequence dissolves in water

Pretty much every DNA/RNA sequence binds its complement

Pretty much every DNA/RNA sequence precipitates in ethanol

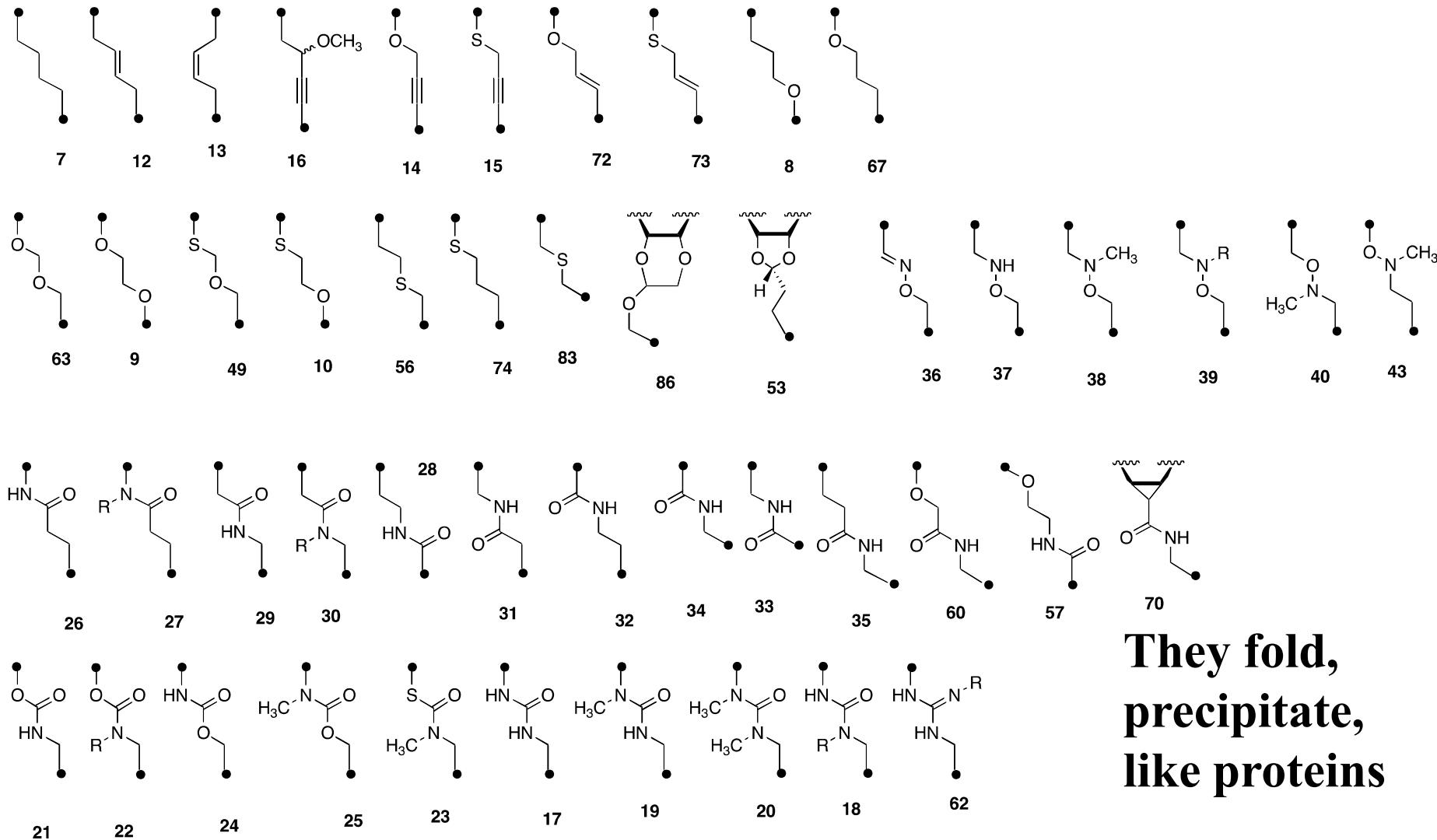
Pretty much every DNA/RNA sequence templates polymerases

What is it about the structure of DNA/RNA ?

**Proteins have a repeating backbone dipole;
DNA has a repeating backbone monopole.**

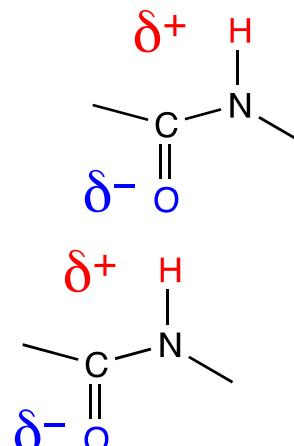
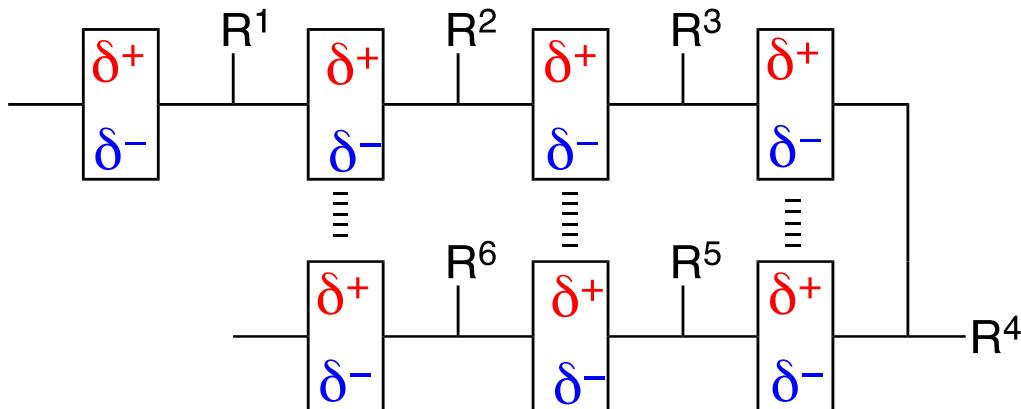
Experimental evidence?

Many DNA analogs without the backbone
charge have been synthesized



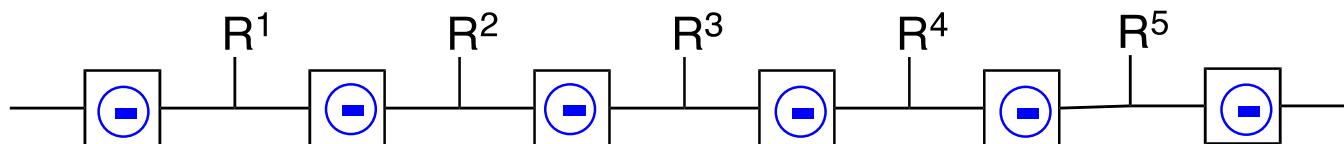
They fold,
precipitate,
like proteins

Compare peptides, with repeating dipole, to DNA/RNA, with a repeating monopole



Think of
magnets
on a
string

(above) A backbone with a repeating dipole easily folds
(below) A backbone with a repeating charge extends to template



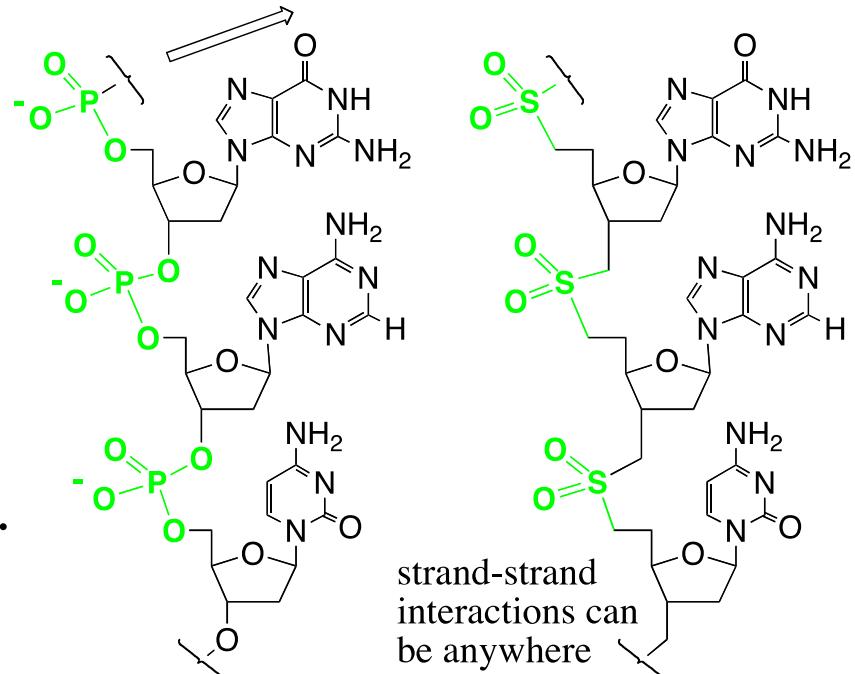
Polyelectrolyte backbone prevents folding, allows templating.
= polyelectrolytes are soluble in water.

Why genetic systems in water must have a polyelectrolyte backbone

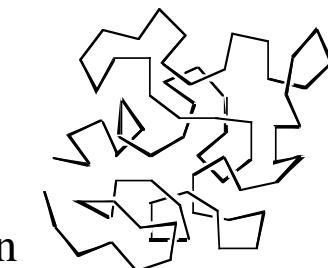
1. Keeps DNA soluble in water.
2. Backbone-backbone coulombic interactions force strand-strand contacts to Watson-Crick edges of the nucleobases (= rules).
3. Repeating charges discourages folding; “excluded volume” effect.
4. Repeating charge dominates the molecule’s properties, allowing mutation to occur without changing the bulk properties of the molecule

Benner, S. A., Hutter, D. (2002) Phosphates, DNA, and the search for nonterrestrial life. Second generation model for genetic molecules. *Bioorg. Chem.* **30**, 62-80

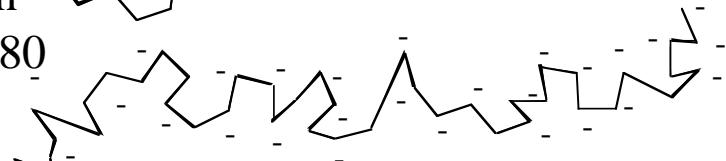
Polyelectrolyte Theory of the Gene
True for all life in water



strand-strand interactions can be anywhere



neutral polymer
Radius = length $(^{1/2})$

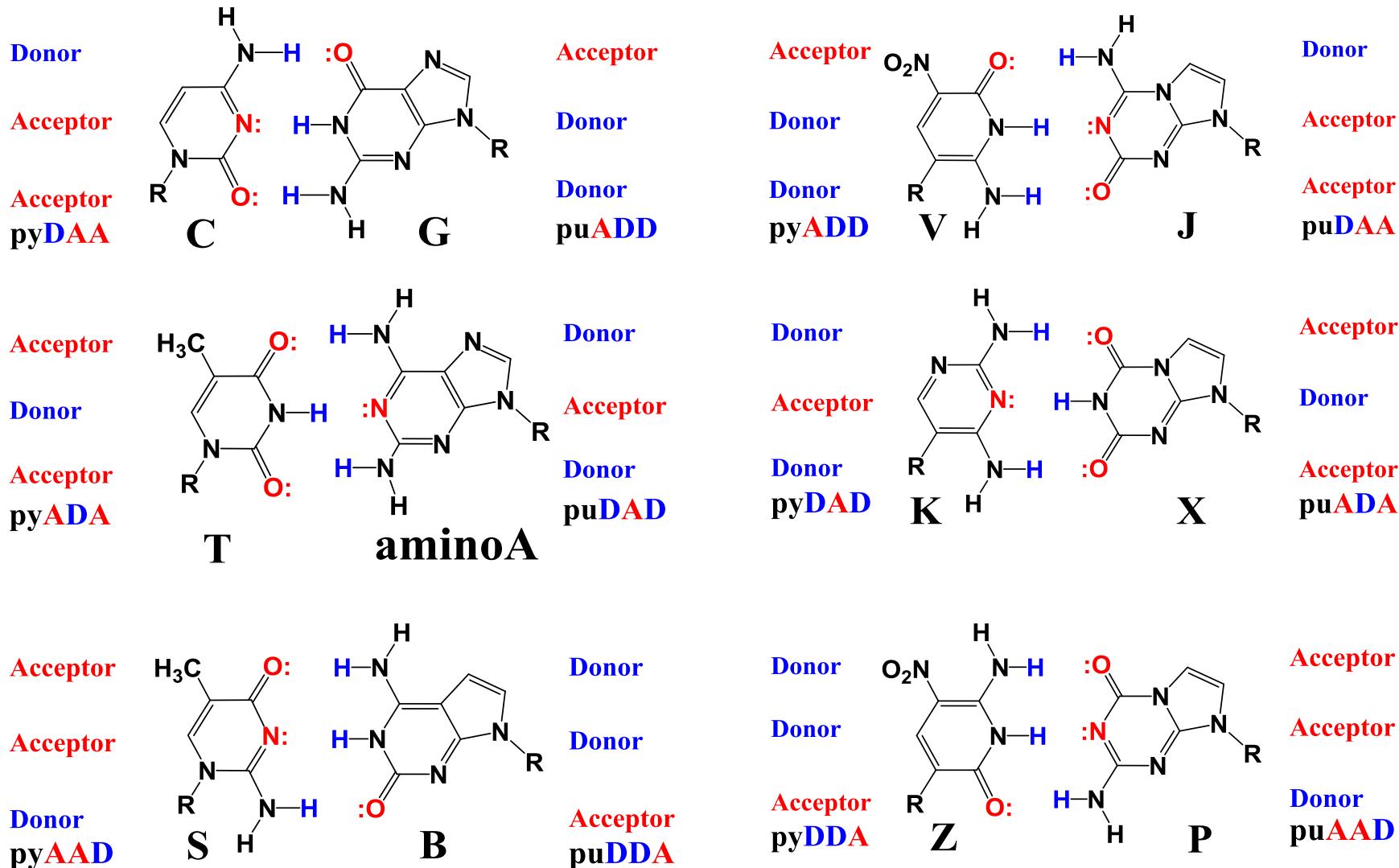


polyanionic polymer
Radius = length $>>(1/2)$



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We can change the bases and retain Darwinism ...



But we cannot change the backbone charges. A universal.

Polyelectrolytes make excellent universal biosignatures

1. Universal genetic biopolymers in water have repeating charges
2. This allows them to be easily concentrated from plumes, bulk water

3. Darwinian biopolymers having this features are easily distinguished

We can today detect extant life in H_2O *in situ*, terran & weird,

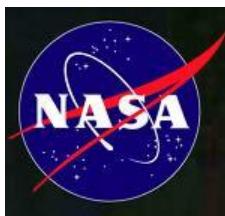
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5. These paths are defeated by dilution into a global ocean
6. Defeat is easily avoided in deserts with occasional water (e.g. Mars)

And non-aqueous life? An analogous universal biosignature?

7. Efforts to get life in cryosolvents defeated by solubility issues
8. Warm Titans, exotic polar solvents, still possible

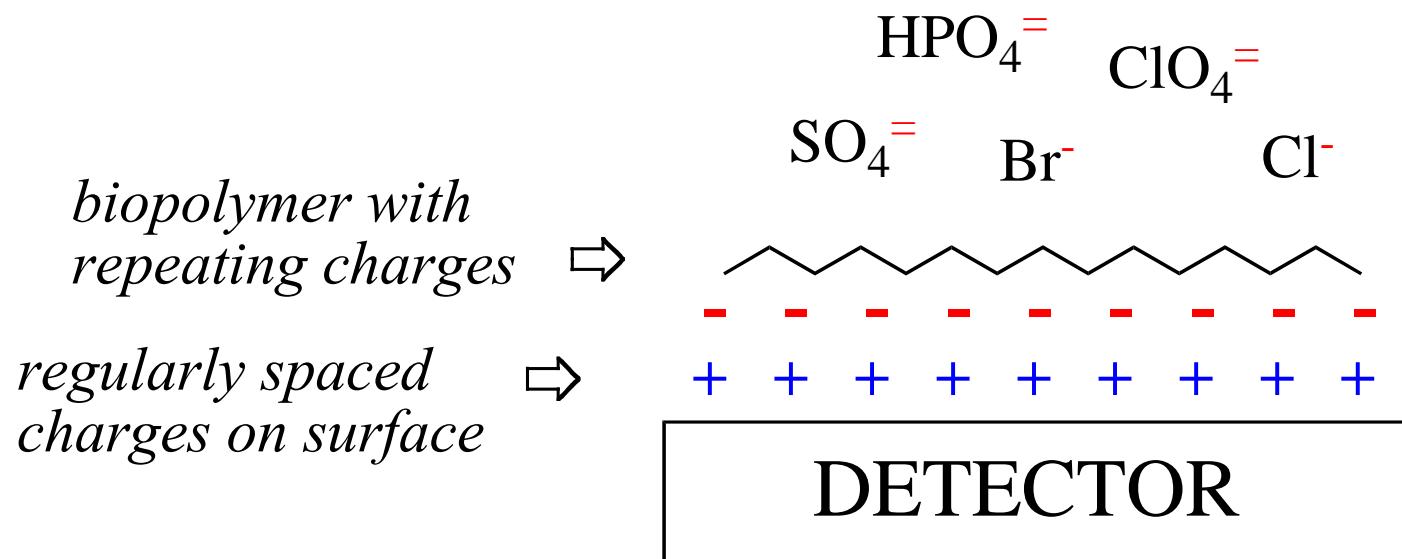
Disequilibrium and the impotence of Darwinism

9. Smart life will escape Darwinism to gain Lamarckianism



Polyanions are easily captured dilute water on polycation supports This is for a *universal* genetic biopolymer

Polyelectrolytes bind in competition with simple ions

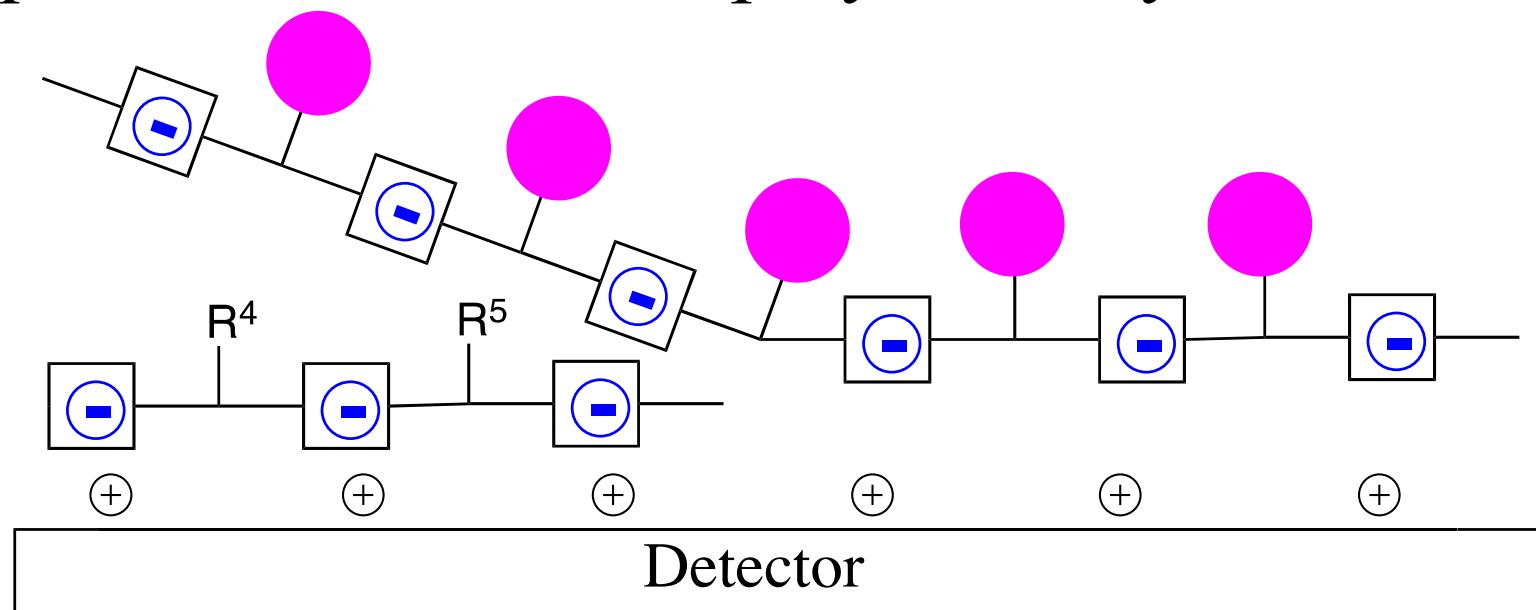


The sample can be indefinitely large and indefinitely dilute; no matter what the specifics of the molecule that support alien genetics, the detector will capture it, and present it to a “detection zone”.



The polyanion is easily detected as part of this capture

Displacement of labeled polyelectrolyte



Sensitivity depends on tags on displaced polyelectrolyte.
Can be radiolabels, fluorescent, UV active, chemically reactive, electrochemically reactive.



Once captured, polyanion easily analyzed to prove that it is Darwinian

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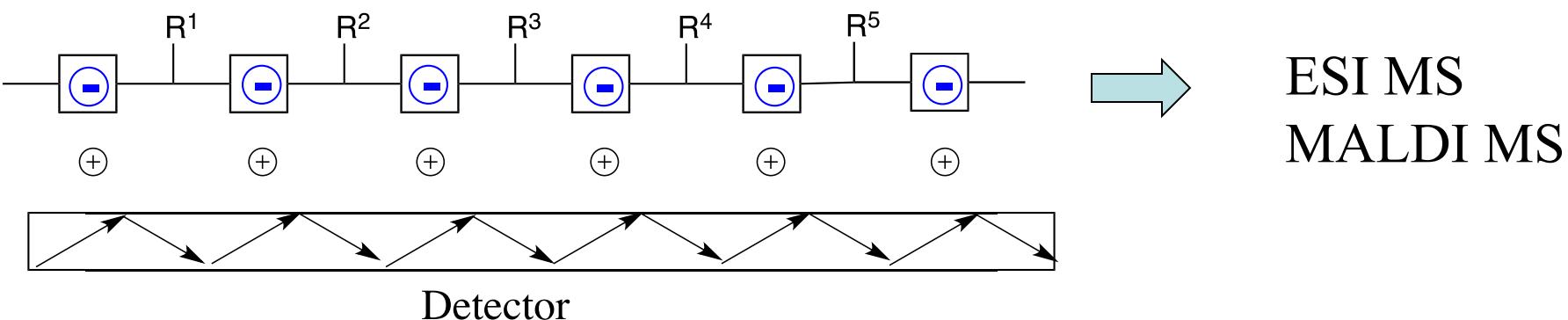
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Darwinian polyelectrolytes are built from a “controlled vocabulary”, a small set of building blocks.

Displace adsorbed polyelectrolyte → Mass spec → fragments

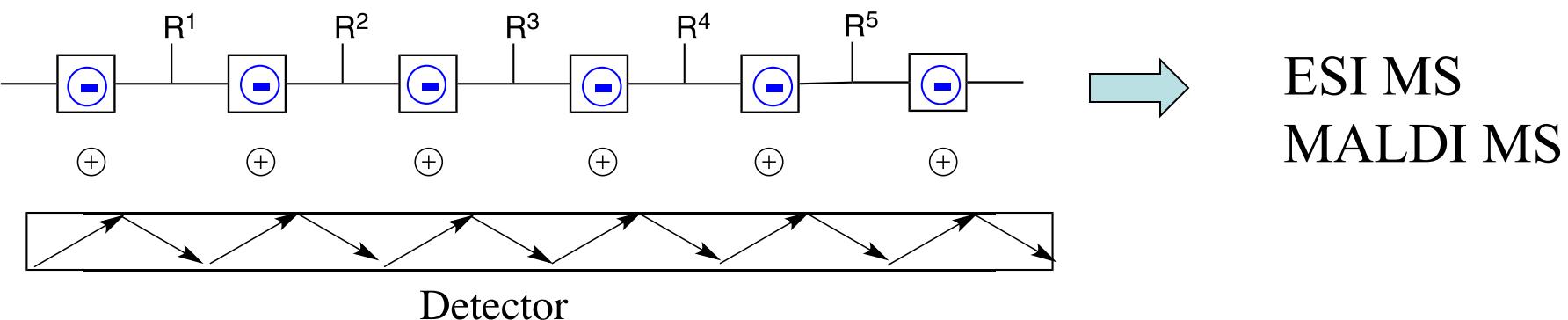


Of course, these must be homochiral to support Darwinism



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Displace adsorbed polyelectrolyte → Mass spec → fragments



Of course, these must be homochiral to support Darwinism

This assay is available today for *in situ* biodetection.

It is universal for any life form in water, anywhere.

Assumes only the universality of Darwinism and the polyelectrolyte theory of the gene.



Which is why I raised my hand to say “yes”. We are ready to go.

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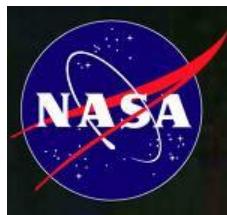
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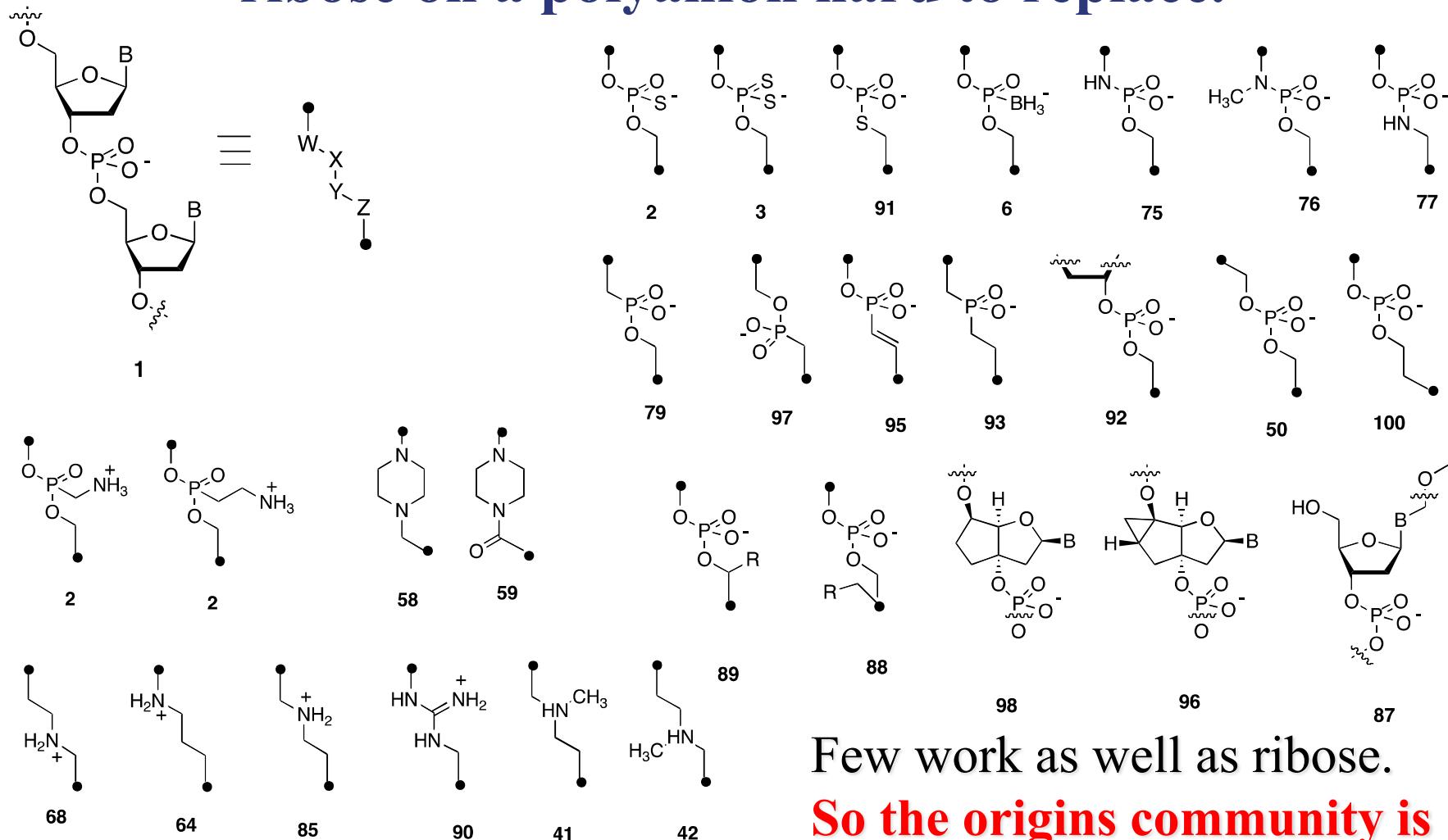
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The rest of genetic biopolymer can be anything. But again, synthesis teaches: ribose on a polyanion hard to replace.



Few work as well as ribose.
So the origins community is
still focused on ribose RNA

We are learning more about how prebiotic RNA might be made

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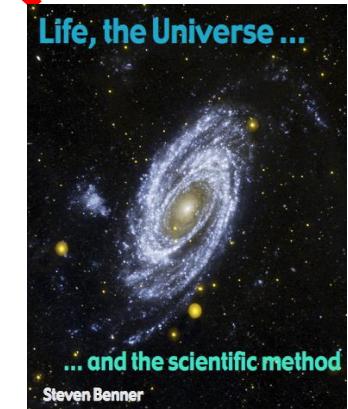
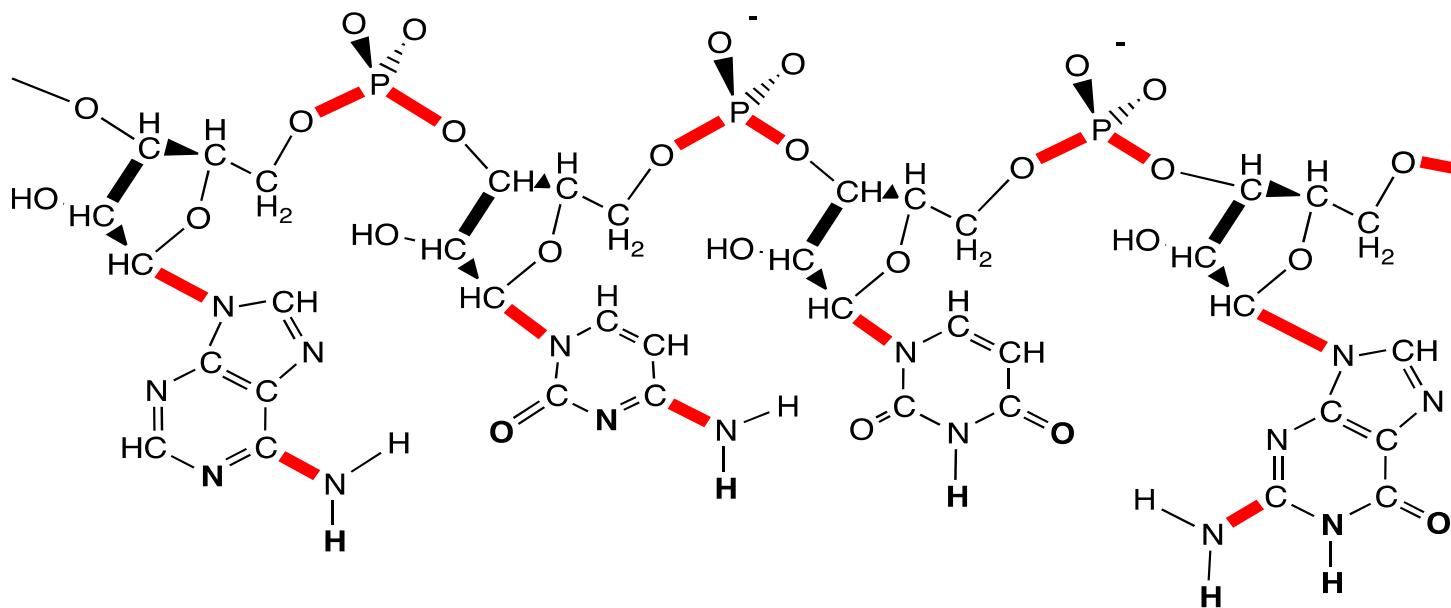
Disequilibrium and the impotence of Darwinism

9. Smart life will escape Darwinism to gain Lamarckianism



I am optimistic because the main problems see solution

- Tar : Give energy to organic matter but no access to Darwinian evolution, one gets **tar**, not **RNA** building units.
Well validated, from the kitchen to the coal field.
- Phosphate. **Hard** to get high phosphate in **RNA**
- **Water**. Even if we get **RNA**, it is **unstable** in **water**.
Water essential but corrosive



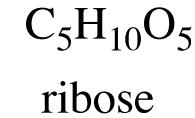
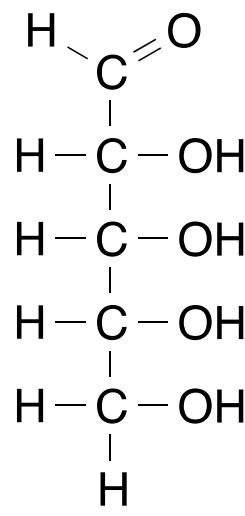
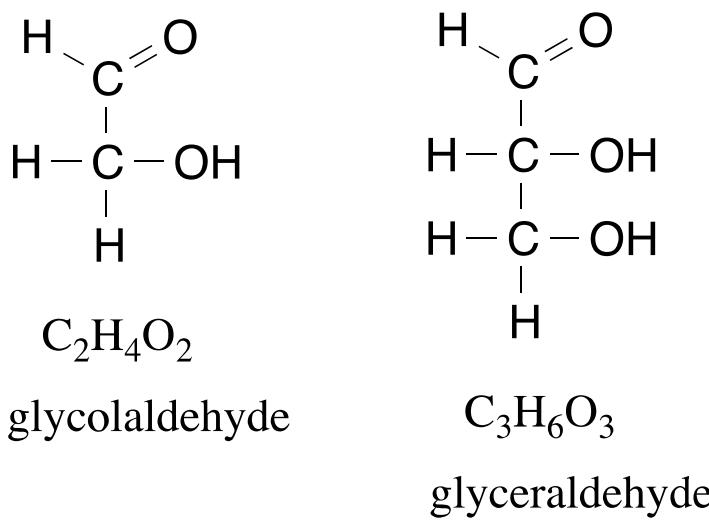
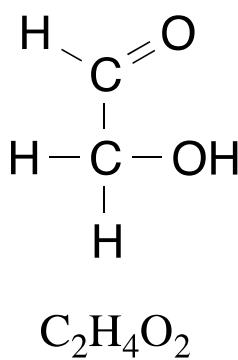
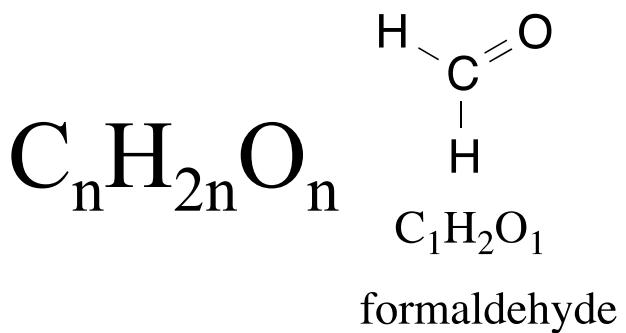
The tar paradox is especially well known with sugars



Sucrose:
"Rock candy"



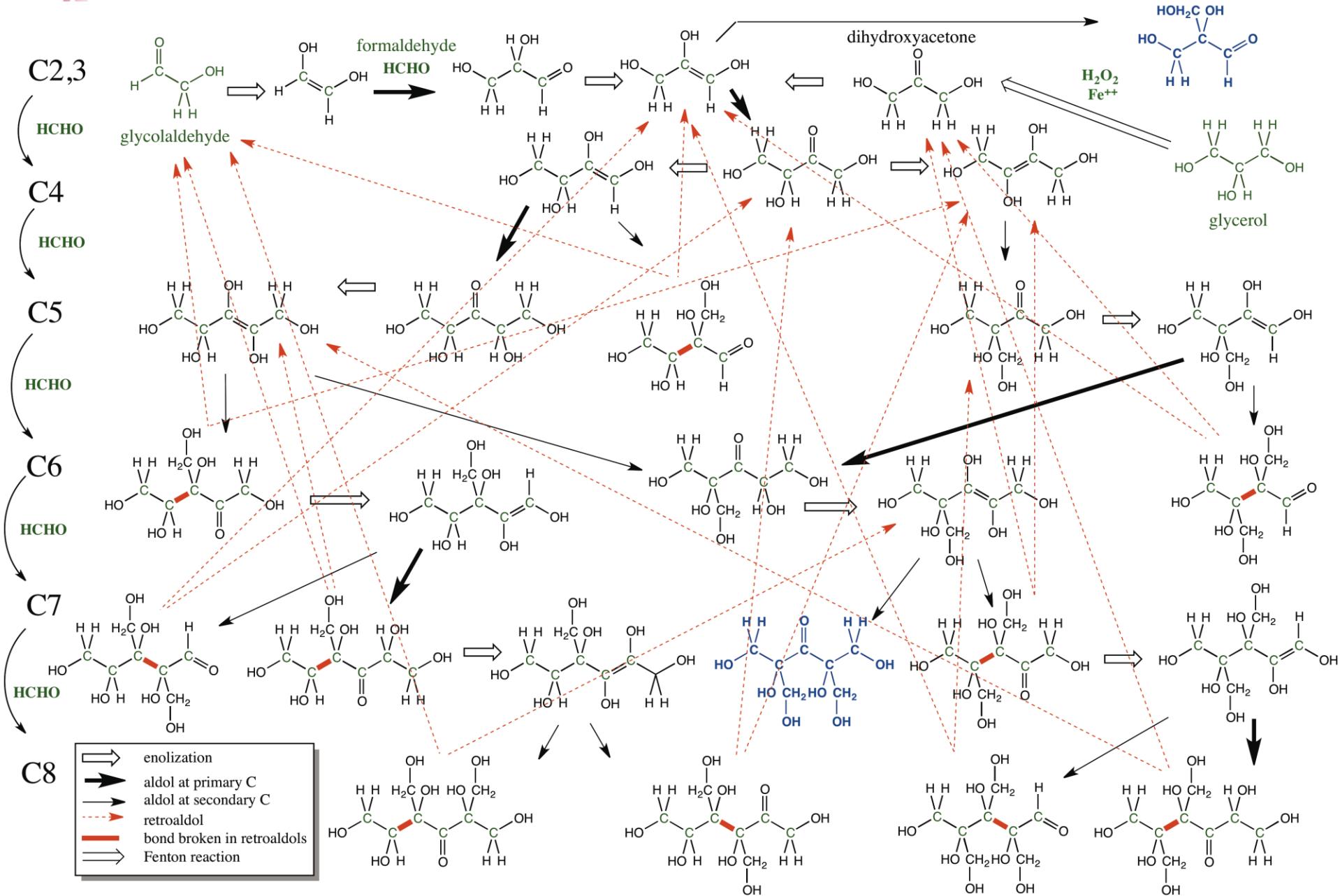
And well known to *you*;
Heat it a bit,
get carmel
Heat more,
get more.



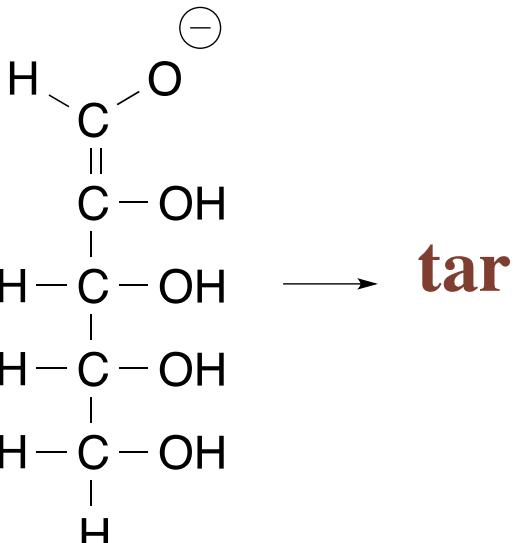
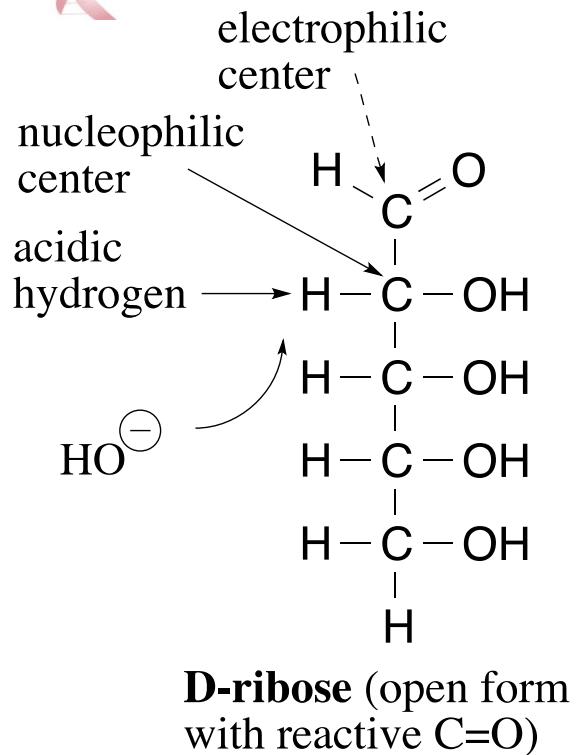
Unfortunately, RNA has a sugar component (R)



Complexity from C=O is horrible



reacts with itself, and forms tar

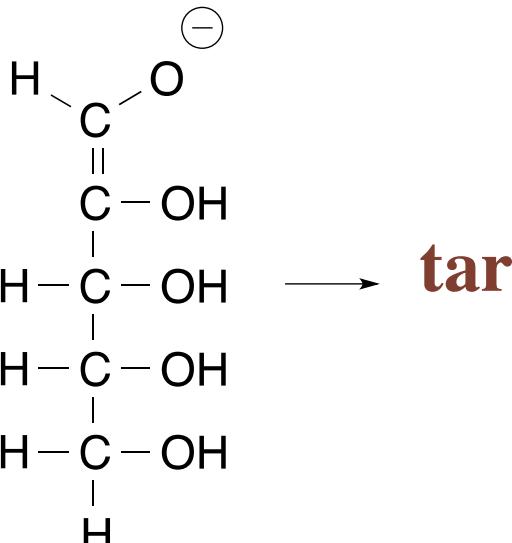
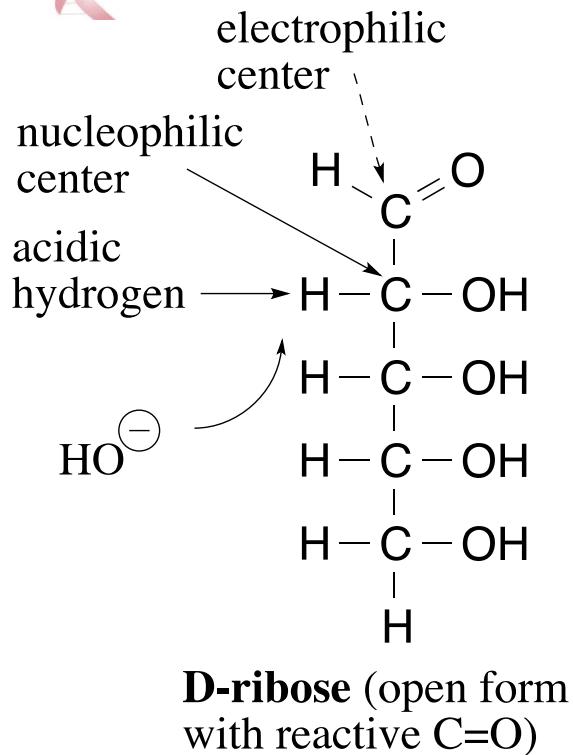


Stanley Miller measured the rate at which ribose formed tar; it is fast (~75 years at pH 7 at 50 °C, not millions of years)

Larralde, Robertson, and Miller (1995) Rates of decomposition of ribose and other sugars. Implications for chemical evolution. *Proc. Natl. Acad. Sci. USA* 92, 8158

"stability considerations preclude the use of ribose and other sugars as prebiotic reagents It follows that ribose and other sugars were not components of the first genetic material..."

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No R = no RNA

We turned mineralogy

Life did not begin in a Pyrex baking pan

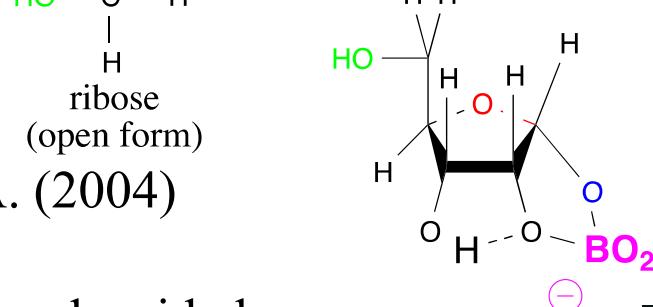
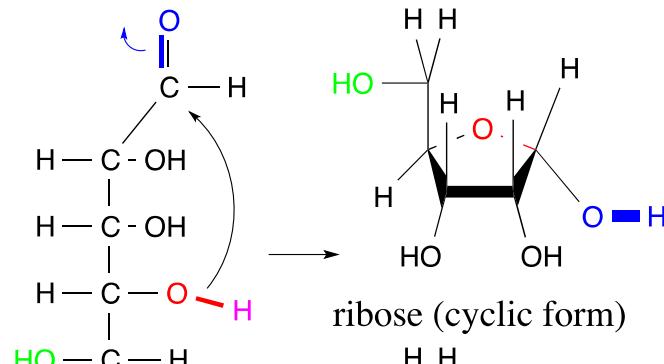
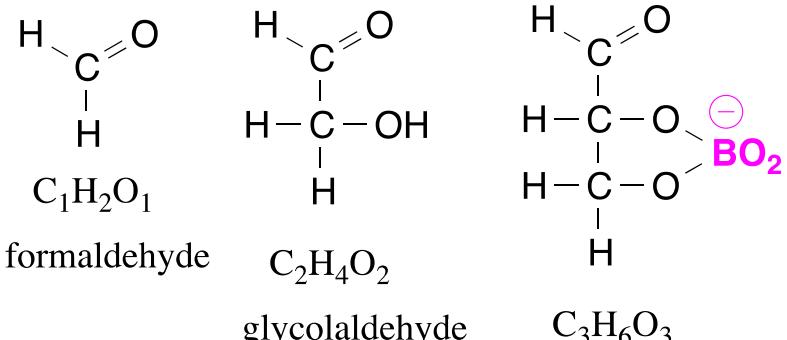
Sugars have adjacent hydroxyl (-OH) groups

Borate binds adjacent hydroxyl groups

Borate binds **ribose**; removes C=O,
prevent tar-ization, allows **ribose** to form.



Borate guides reaction of carbohydrates

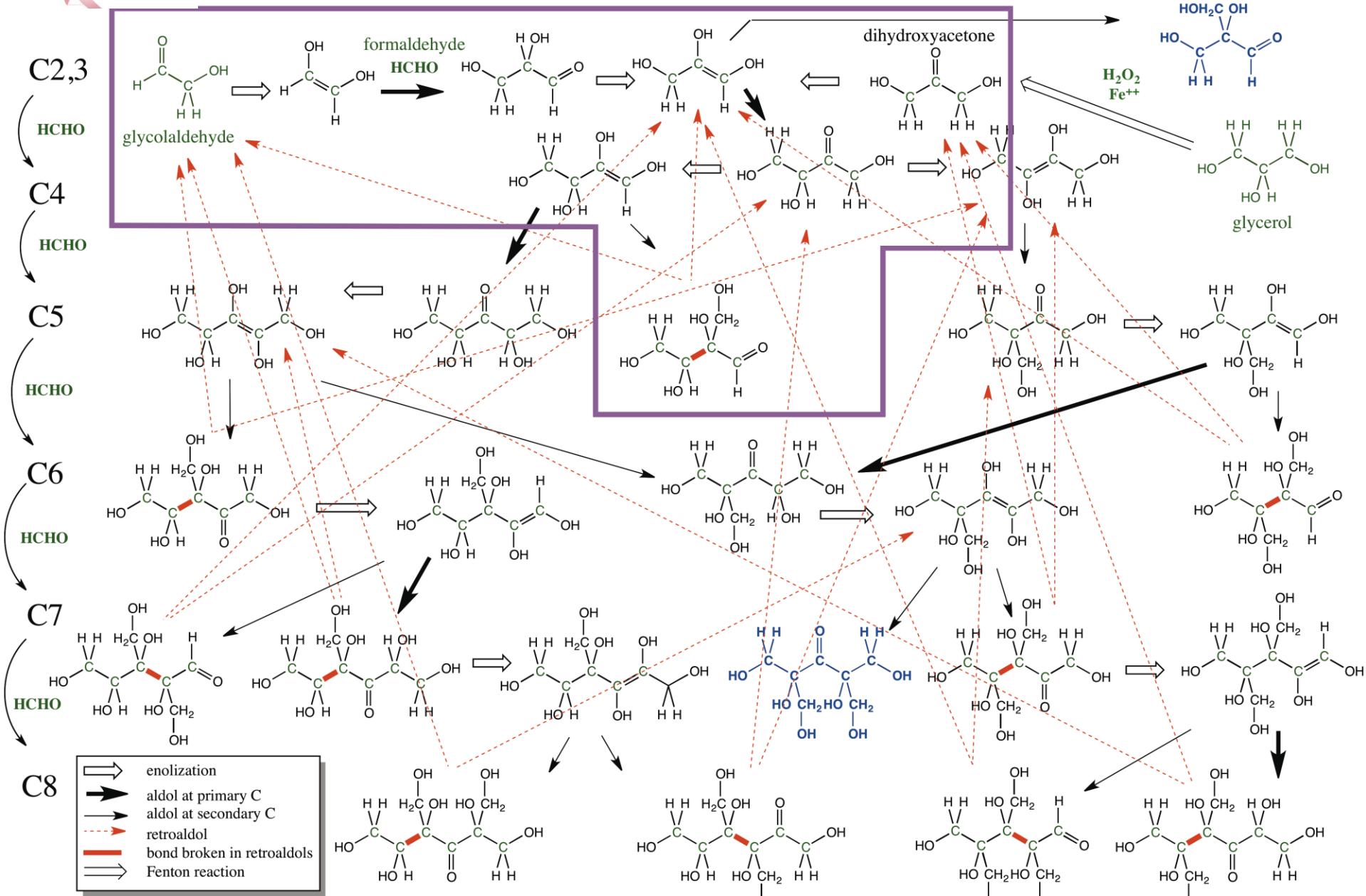


Ricardo, A., Carrigan, M.A., Olcott, A., Benner, S.A. (2004)

Borate minerals stabilize **ribose**. *Science* **303**, 196

Kim, et al. (2011) Synthesis of carbohydrates in mineral-guided prebiotic cycles. *J. Am. Chem. Soc.* **133**, 9457-9468

Borate constrains reactivity, makes ribose



Desert environments where borate minerals accumulate also have low water activity.



Peridot in basalt
generates the base



Tourmaline (borate)

Borate excluded from silicates, comes to surface in igneous rocks, easily weathered, most salts water soluble, collect in dry basins. Peridot in basalt weathers to creates alkali.

--> *Borate moderated formose*

Wulfenite (molybdate)



Evaporite minerals
saturated borate
Colemanite= boron, calcium, oxygen, hydrogen



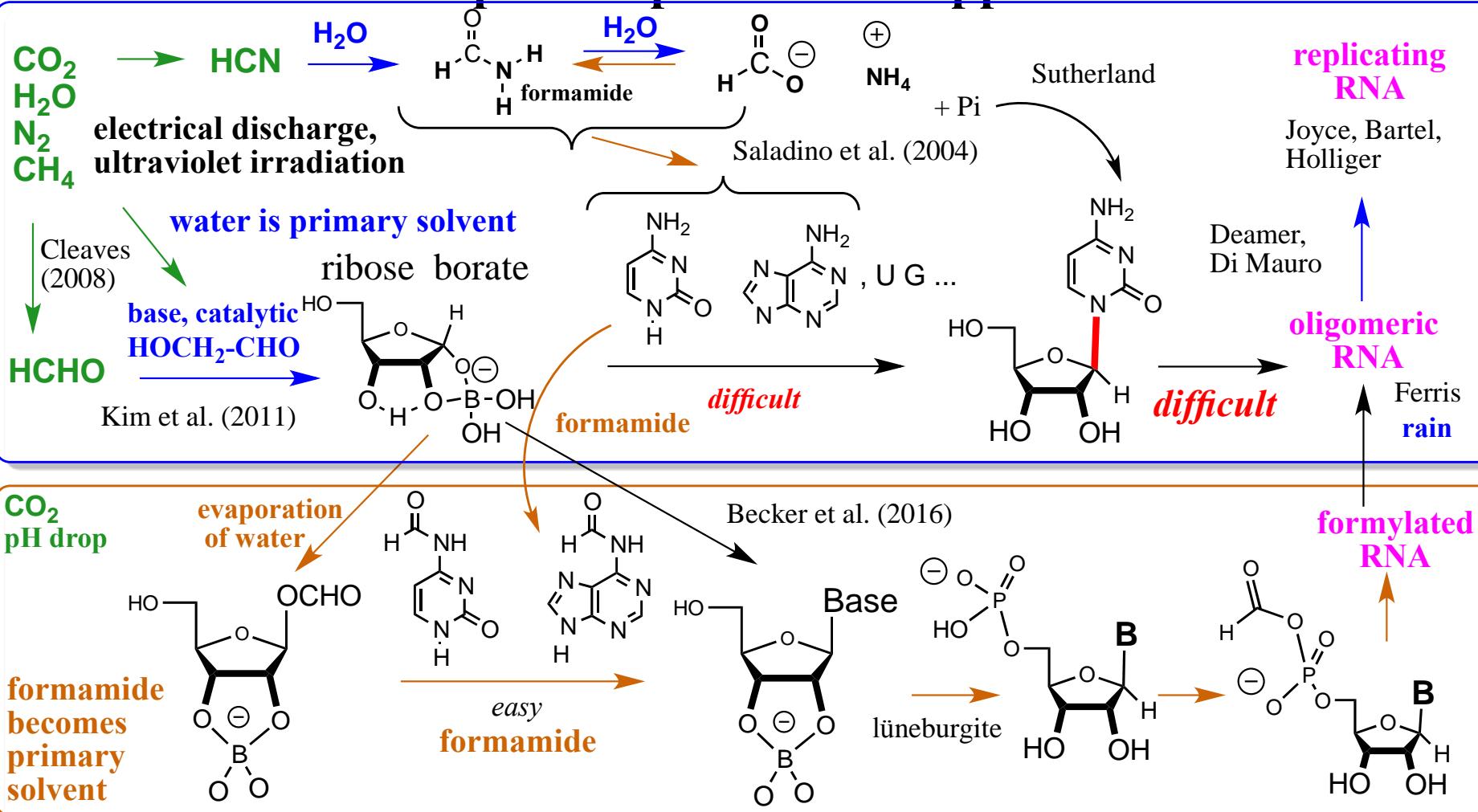
Death Valley

Origin of life jewelry

Discontinuous Model for RNA synthesis

I think the solution will look something like this.

Each step has experimental support



Key: in the atmosphere

in the aquifer; Nucleophilic Context

in desert evaporite; Electrophilic Context



needs intermittent water

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Were these minerals and deserts present on early Earth?



Molybdates are highly oxidized



tourmaline

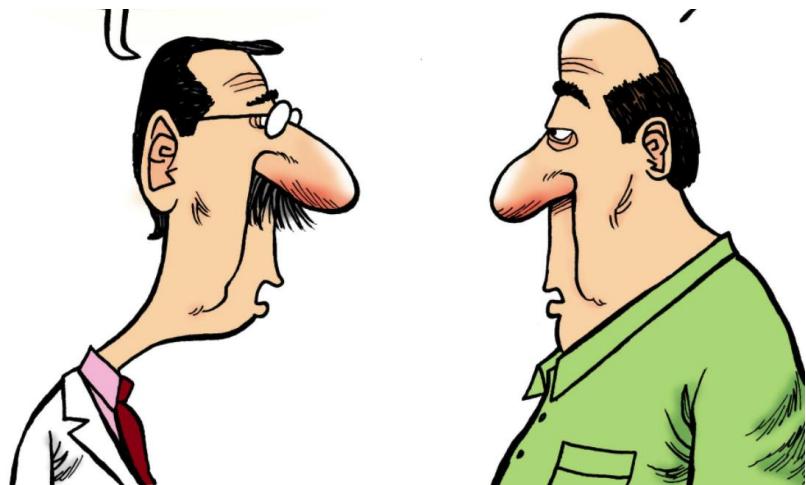


colemanite

Early Earth borate? Hazen and Grew say “no”.
Early Earth evaporites? Kirschvink says “no”.

*What can you give me by way
of minerals on early Earth?*

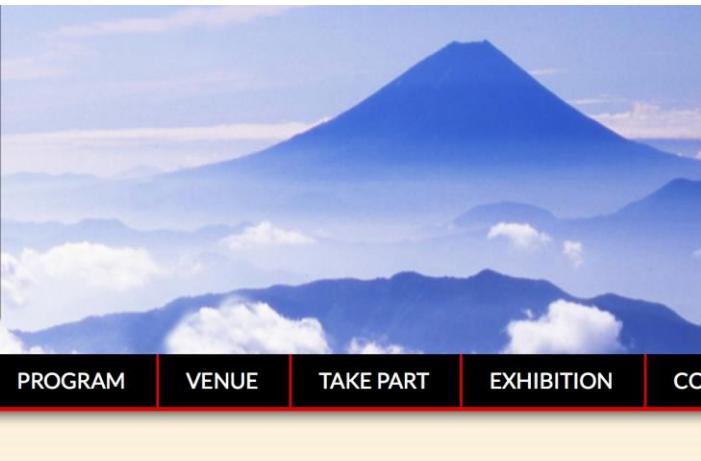
What do you need?



Tourmalines, colemanite, and other borate minerals are formed only if scarce boron is enriched. In the lithosphere, only after some cycling time? In the hydrosphere, only in deserts.

But our work has caused geologists to look again

If geologists *look* for Archaean borate ... They find it



Tourmaline-Bearing Crusts in the 3.48 Ga Dresser Formation: Derivation and Implications for the Origin of Life

[Van Kranendonk M](#), Ota T, Nakamura E & Djokic T

Borate minerals unstable to metamorphism, give monazite, apatite, and tourmaline. These are in ~3.8 Ga metasedimentary rocks from the Isua supracrustal belt (Greenland) with biogenic (?) graphite.

Mishima, S., Ohtomo, Y., Kakegawa, T. (2016) Occurrence of tourmaline in metasedimentary rocks of the Isua supracrustal belt, Greenland. Implications for ribose stabilization in Hadean Marine Sediments. *Origin Life Evol. Biosphere* **46**, 247-271.

Grew, E. S. et al., (2015) Boron isotopes in tourmaline from the ca. 3.7-3.8 Ga Isua supracrustal belt, Greenland: Sources for boron in Eoarchean continental crust and seawater. *Geochim. Cosmochim. Acta*. **163**, 156-177.



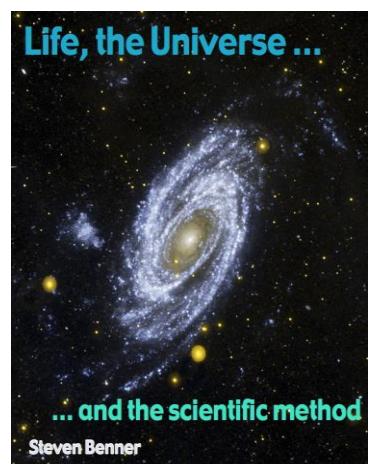
Are deserts available on early Earth?

Joe Kirschvink's objection to the Benner model. Models for planetary formation suggest that the inventory of water on early Earth did not leave *any* dry land before continental drift.

No deserts, no borate evaporites, no ribose, no R, no RNA.



Waterworld



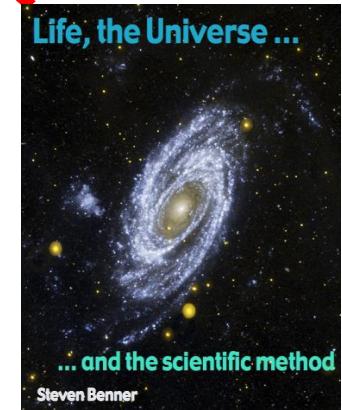
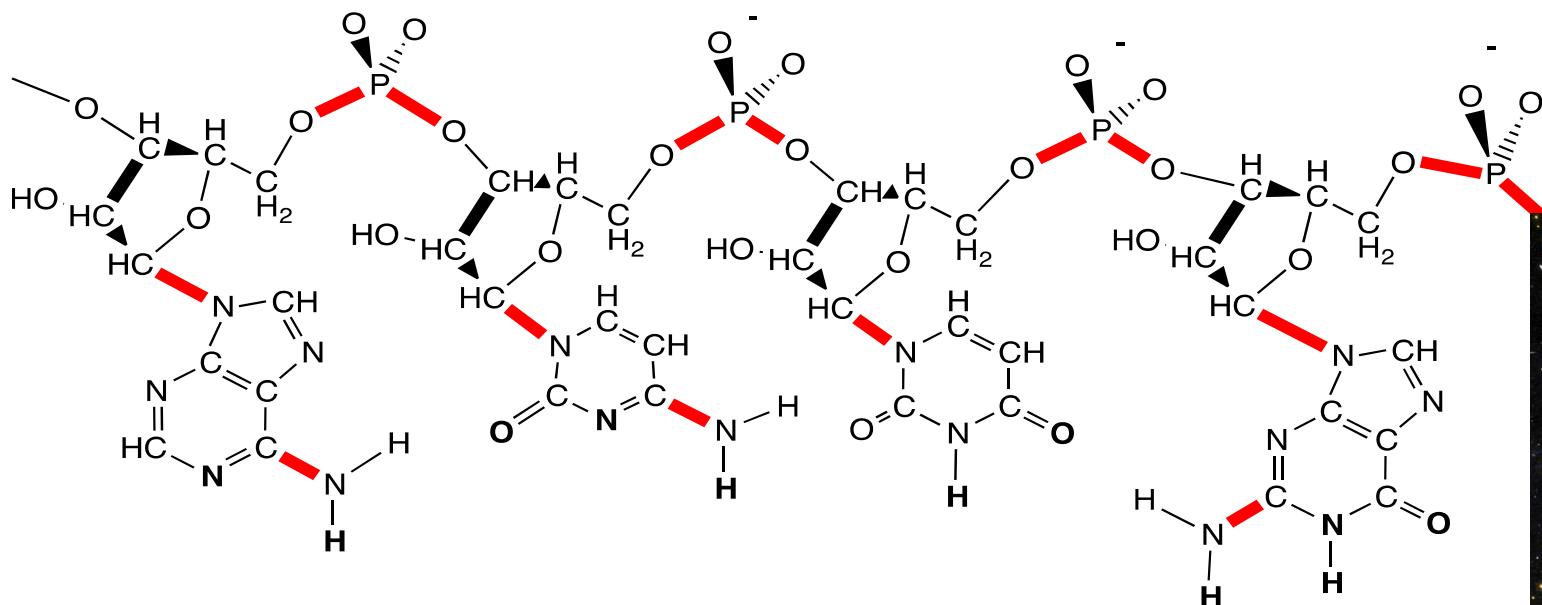
The Kirschvink Modification of the Benner Model

Move it to Mars, where water was never as abundant and oxidation level has always been higher. *Perhaps borate-ribose is there today.*



I am optimistic because the main problems see solution

- Tar : Give energy to organic matter but no access to Darwinian evolution, one gets **tar**, not **RNA** building units.
Well validated, from the kitchen to the coal field.
- Phosphate. **Hard** to get high phosphate in **RNA**
- **Water**. Even if we get **RNA**, it is **unstable** in **water**.
Water essential but corrosive



Borate +phosphate = borophosphate minerals, not inaccessible apatite

27. LÜNEBURGITE [$\text{Mg}_3(\text{PO}_4)_2\text{B}_2\text{O}(\text{OH})_4 \times 6\text{H}_2\text{O}$] IN UPPER MIOCENE SEDIMENTS OF THE EASTERN MEDITERRANEAN SEA

Jens Müller and Frank Fabricius, Lehrstuhl für Geologie, Technische Universität, München, West Germany

Lüneburgite



ABSTRACT

Upper Miocene dolomitic marls drilled at Site 374 (Messina Abyssal Plain, Ionian Sea) contain small white mm-sized spherules which have been identified by X-ray, chemical, and IR-spectrometry techniques as lüneburgite $[\text{Mg}_3(\text{PO}_4)_2\text{B}_2\text{O}(\text{OH})_4 \times 6\text{H}_2\text{O}]$.

We assume that the lüneburgite formed as a secondary product by upward migrating brines from the Miocene evaporites which contain highly soluble Mg- and B- salts. Enrichment of phosphorus may be the result of ~~concentration in residual brines from the~~ chiefly of authigenic Ca-dolomite ($\text{Ca}_{55}\text{Mg}_{45}$) and the detrital components quartz, feldspar, smectite, illite, kaolinite, chlorite, and traces of (?)attapulgite. The fine-grained black sediment is slightly enriched in organic carbon (0.3-0.7%; Sigl, this volume) and when fresh, expells a bituminous odor. Secondary(?) gypsum occurs within the sequence containing lüneburgite. These homogeneous sediments were deposited as a transitional facies between the formation of evaporites and deposition of more open marine sediments (Site 374 Report, this volume).

Tribal jargon

Gypsum = $\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$

Lüneburgite =

$\text{Mg}_3\text{B}_2(\text{PO}_4)_2(\text{OH})_6 \cdot 8\text{H}_2\text{O}$

What does it mean if gypsum + lüneburgite occur together?

The phosphate problem in the “RNA first” model

Clearly stoichiometric phosphate is necessary for RNA itself

Powner et al. (200) need it in quantity (1 M, 98 g/L)

But phosphate + calcium precipitate = apatite

Ca^{+2}

PO_4^{-3}



apatite

SO_4^{-2}

Mg^{+2}



epsomite



Apatite

What happens with borate?



Supported by a
grant from

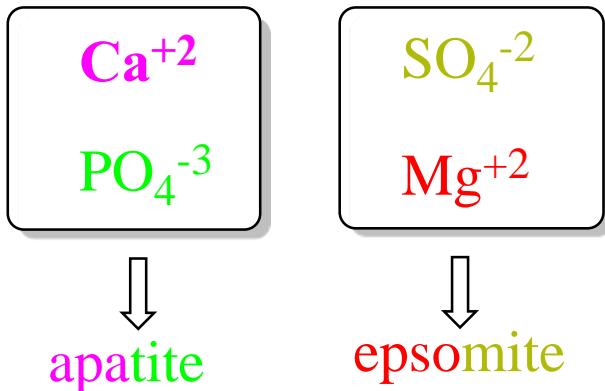
John
Templeton
Foundation

Benner collection of fine minerals

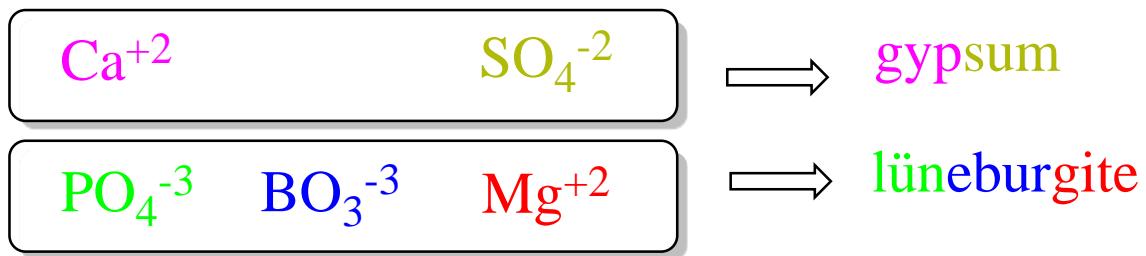
Borate keeps phosphate away from apatite

In natural environments, shown by observational geology

Partitioning of species
in absence of **borate**



Partitioning of species
in presence of **borate**

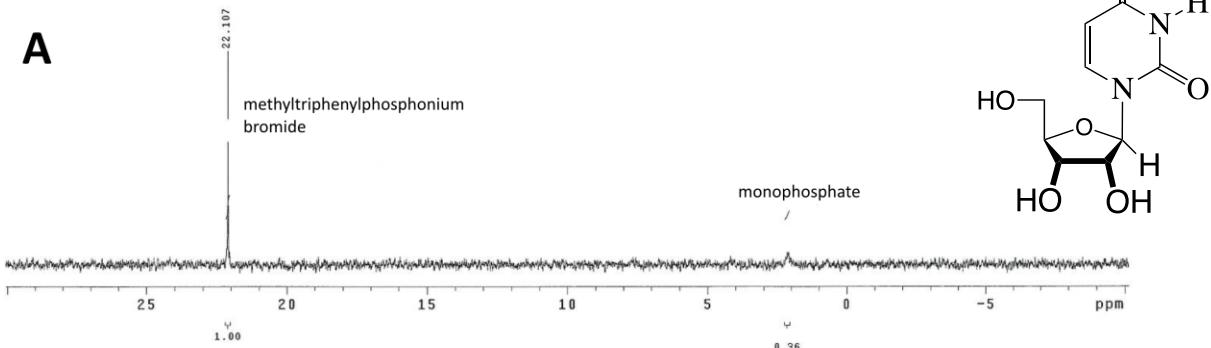


This also happens in the laboratory if you mix MgO , CaO , H_3BO_3 , H_3PO_4 , and H_2SO_4 and evaporating.

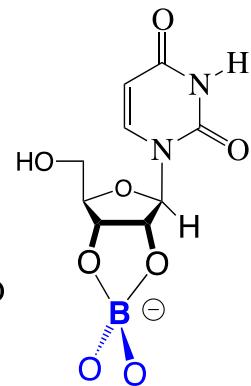
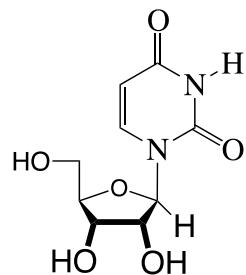
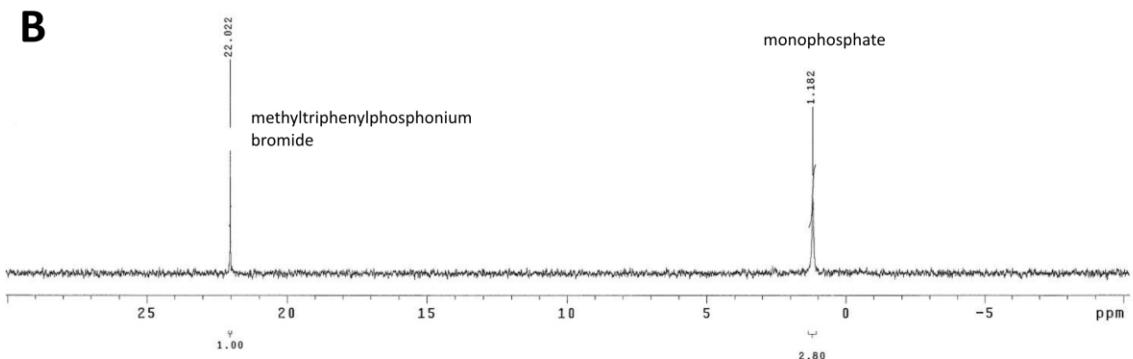
**Borate keeps phosphate from being lost by calcium capture.
Borophosphate formed instead.**

Ribose extracts borate out of luneburgite, makes phosphate available for phosphorylation

A



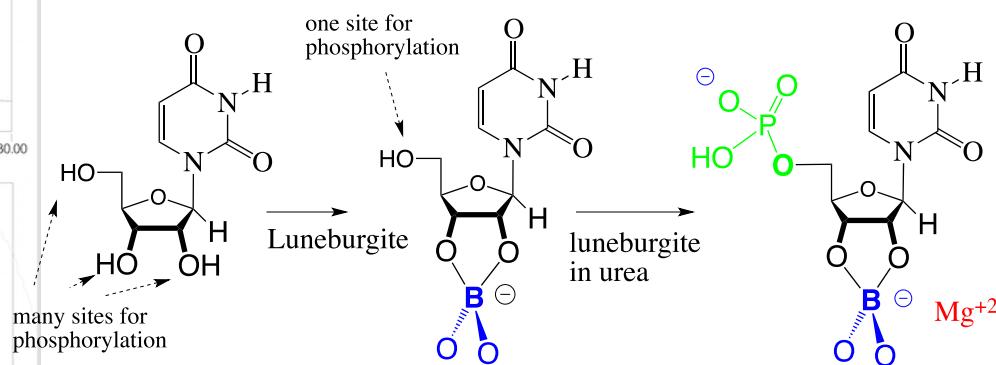
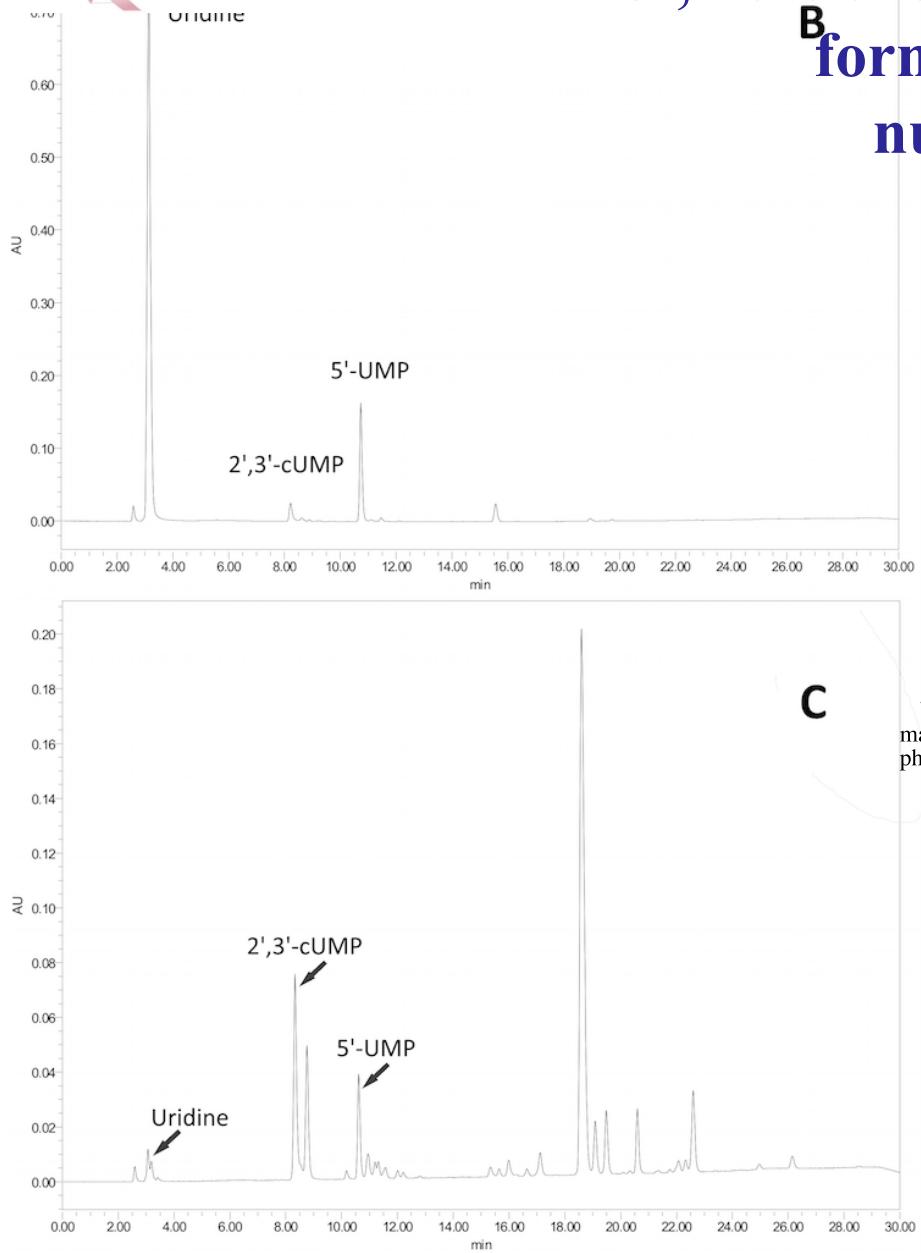
B



Kim, H. J., Furukawa, Y., Kakegawa, T., Bita, A., Scorei, R., Benner, S. A. (2016) Evaporite borate-containing mineral ensembles make phosphate available and regiospecifically phosphorylate ribonucleosides: Borate as a multifaceted problem solver in prebiotic chemistry. *Angew. Chem.* DOI: 10.1002/ange.201608001

In non-aqueous media (formamide or, here urea) where phosphate ester formation is thermodynamic, nucleosides regioselectively phosphorylated.

Absent borate, one gets a mixture.



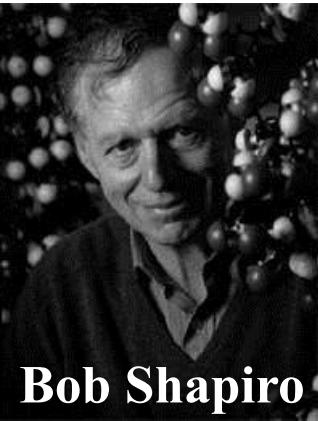
Two for the price of one.

Lüneburgite sequesters phosphate from calcium until nucleosides are present, then phosphorylates nucleosides regioselectively after nucleosides decompose the mineral to release phosphate.

Where do nucleosides come from?

Pownier and Sutherland have a route Thomas Carell et al. have a better route

Carell route starts with ribose **borate**, *Unstable carbohydrates (glyceraldehyde) need not accumulate for Carell's route to work.*

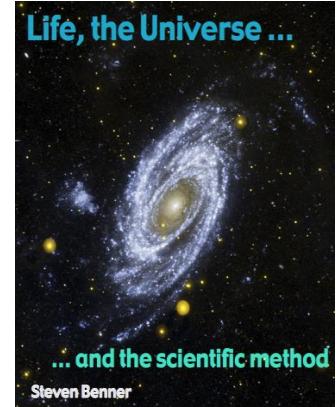


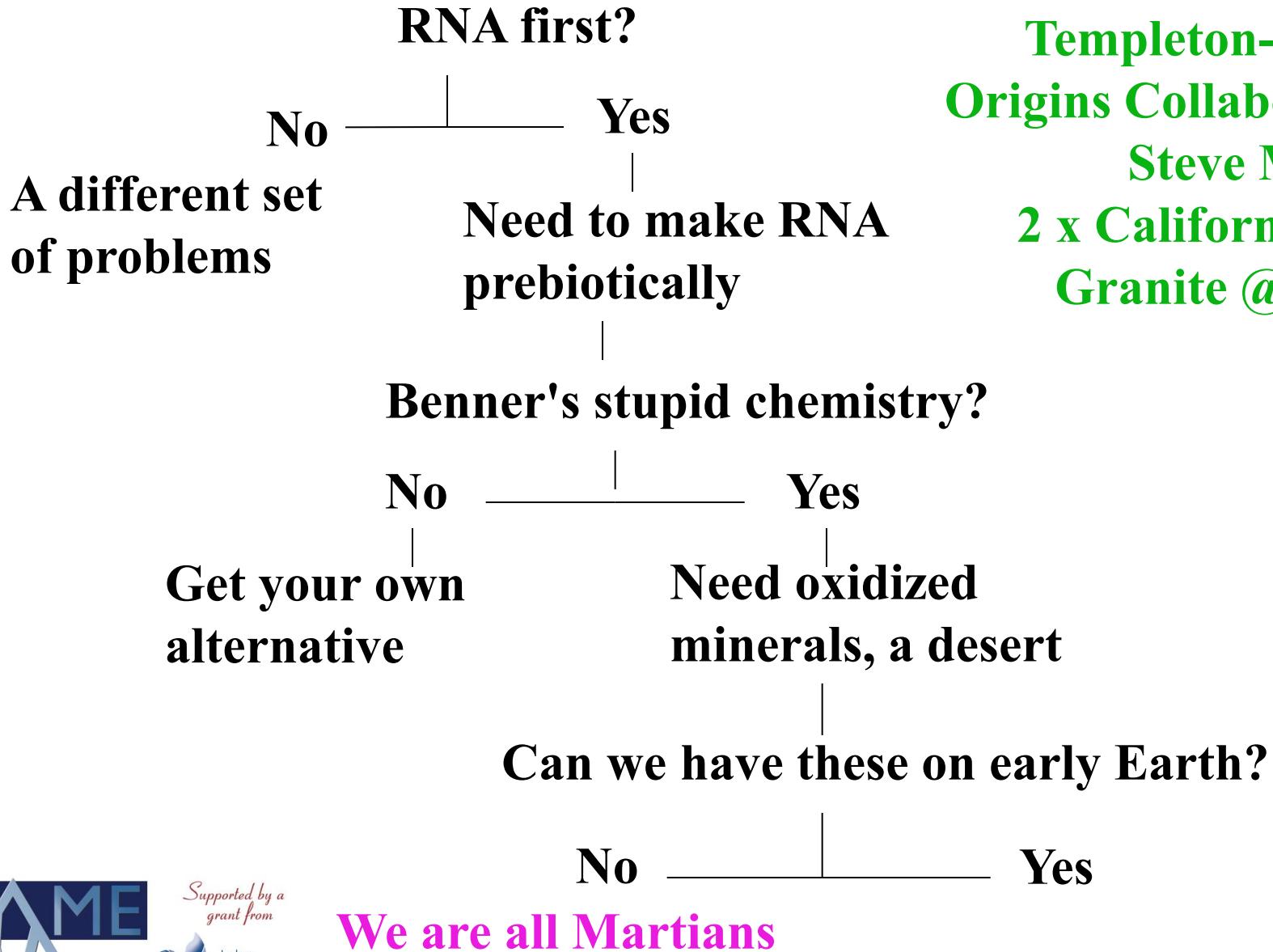
I am optimistic because the main problems see solution

- Tar : Give energy to organic matter but no access to Darwinian evolution, one gets **tar**, not **RNA** building units.
Well validated, from the kitchen to the coal field.
- Phosphate. **Hard** to get high **phosphate** in **RNA**
- **Water**. Even if we get **RNA**, it is **unstable** in **water**.
Once RNA formed, adsorbed onto CT-Opal



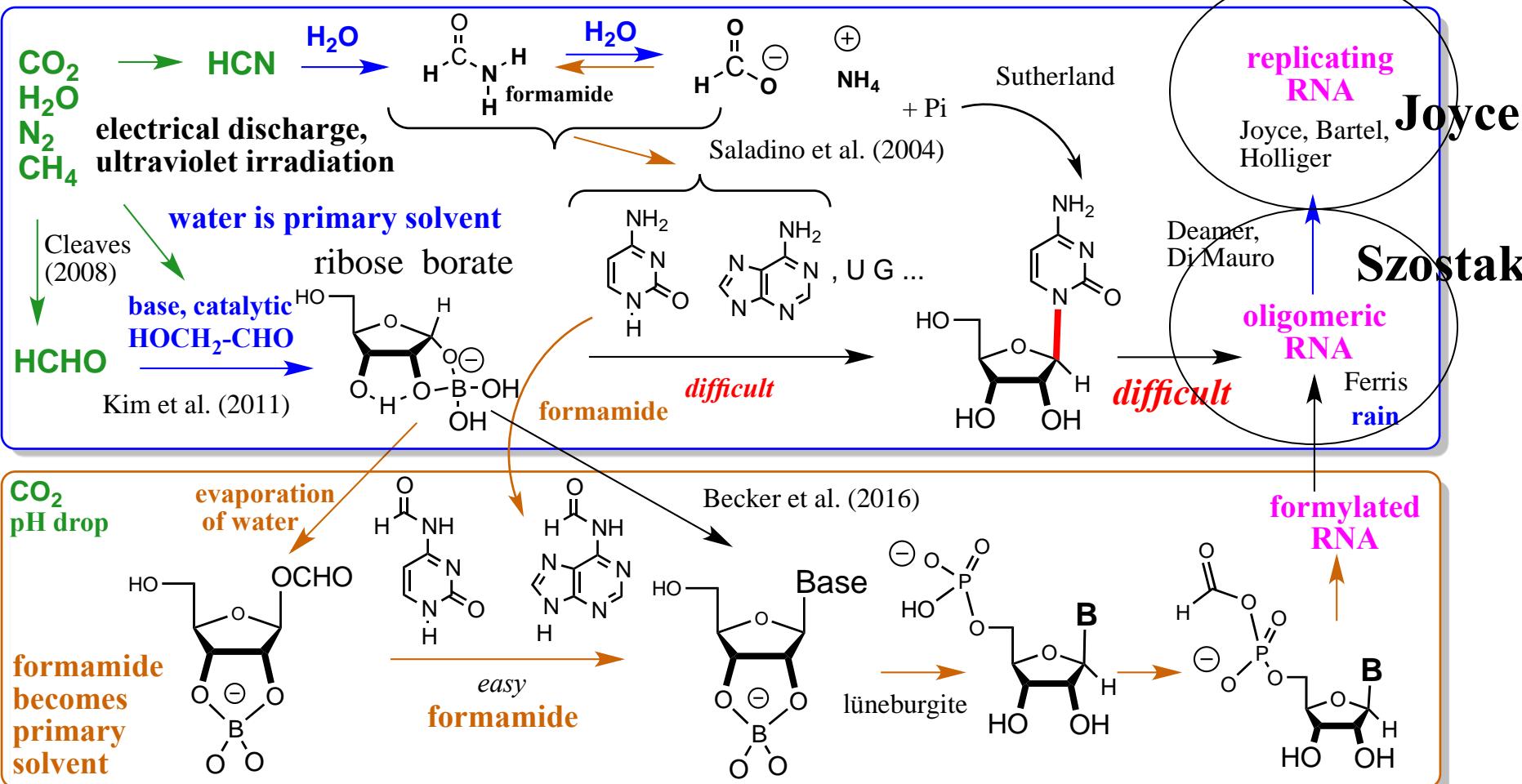
John Grotzinger, David Blake, **exactly like 3 Ga Mars**





Model for prebiotic synthesis of RNA

Given existing experimental support, a likely schema



There is hope, even though problems remain

Aqueous life needs dry land. Perpetual water worlds a problem.

1. Universal genetic biopolymers in water have repeating charges
2. This allows them to be easily concentrated from plumes, bulk water
3. Darwinian biopolymers having this features are easily distinguished

We can today detect extant life in H_2O *in situ*, terran & weird,

4. We still lack a clear path to get RNA as the first terran Darwinian biopolymer, but it seems that we *will* find those paths.

5. These paths are defeated by dilution into a global ocean
6. Defeat is easily avoided in deserts with occasional water (e.g. Mars)

And non-aqueous life? An analogous universal biosignature?

7. Efforts to get life in cryosolvents defeated by solubility issues
8. Warm Titans, exotic polar solvents, still possible

Disequilibrium and the impotence of Darwinism

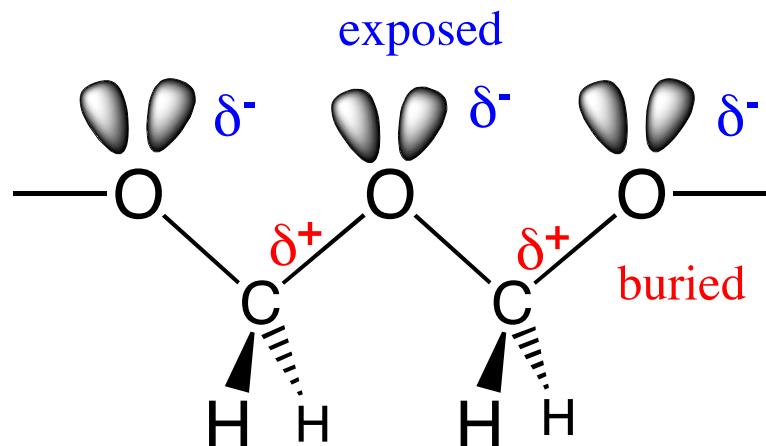
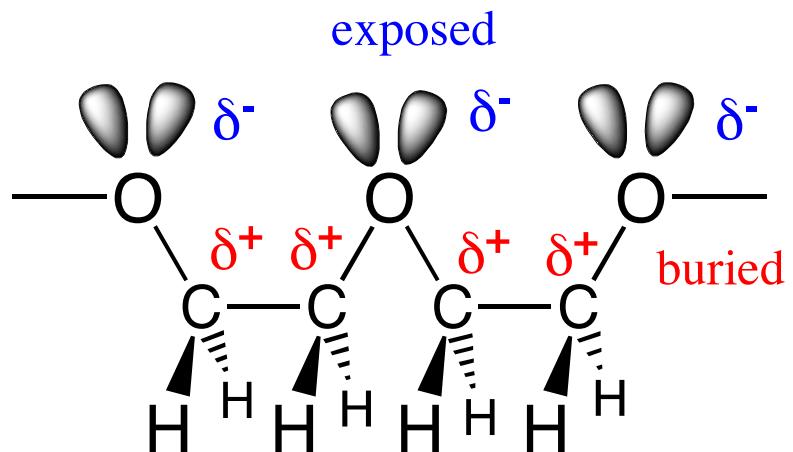
9. Smart life will escape Darwinism to gain Lamarckianism



Biopolymer with a repeating charge will not dissolve in hydrophobic solvents.

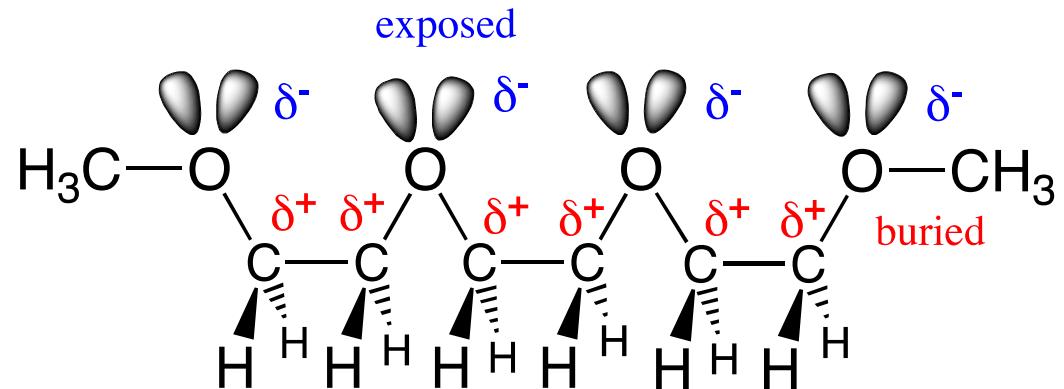
What about a biopolymer with a dipole, where the exposed part of the dipole is negatively charged?

By burying consistently one end of the dipole, and exposing the other, maybe the polymer cannot aggregate.



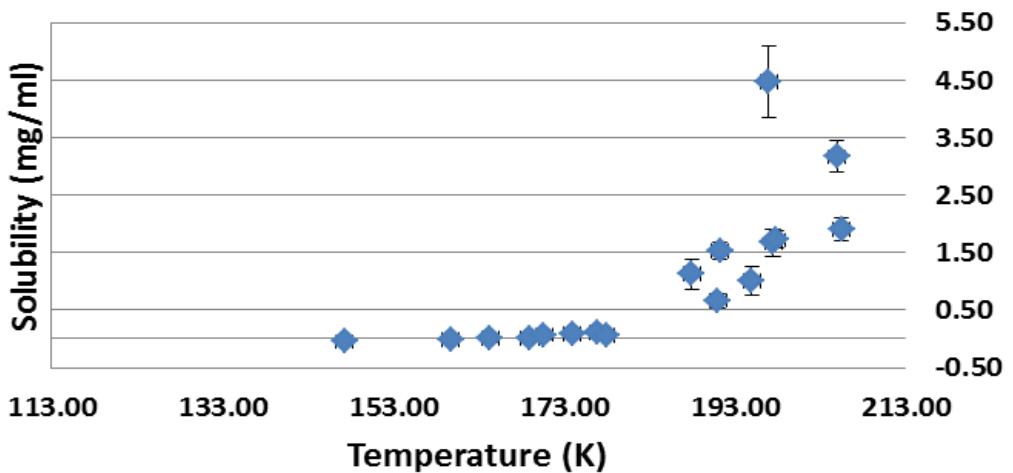
McLendon, C., Opalko, F. J., Illankoon, H. I., Benner, S. A. (2015) Solubility of polyethers in hydrocarbons at low temperatures. A model for potential genetic backbones on warm Titans. *Astrobiol.* **15**, 200-206.

They dissolve in warm hydrocarbons; aggregate (precipitate) in cold



Hydrocarbon	Freeze	Boil
Methane	91 K	112 K
Ethane	89 K	184 K
Propane	85 K	231 K
Butane	135 K	273 K

$\text{C}_8\text{H}_{18}\text{O}_4$ Tri-ethylene glycol dimethyl ether
(4 assays)



A "warm Titan" is OK.
Water is a good solvent because it is hot.
William Bains



Cryosolvents are not good, if you think that biology needs dissolution

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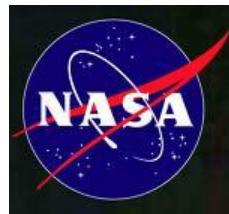
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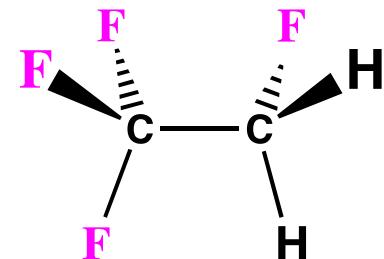
A disequilibrium that shows the impotence of Darwinism.
It has not created a life form able to exploit the energy in the tree-dioxygen combination. The solution?

The solution: Lamarckianism



Homo
erectus
invented
Lamarckian
evolution

Any life form smart enough to talk to with us will soon get control of its biology to not need dying babies die to prevent sickle cell anemia. *Germ line gene therapy*. Universal for life, regardless of core molecular biology. **What biosignatures would come from this life form?**



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Search for Lamarckian biosignatures escapes conundrum of remote detection of unknown molecular biology.

