The molecular origins of life

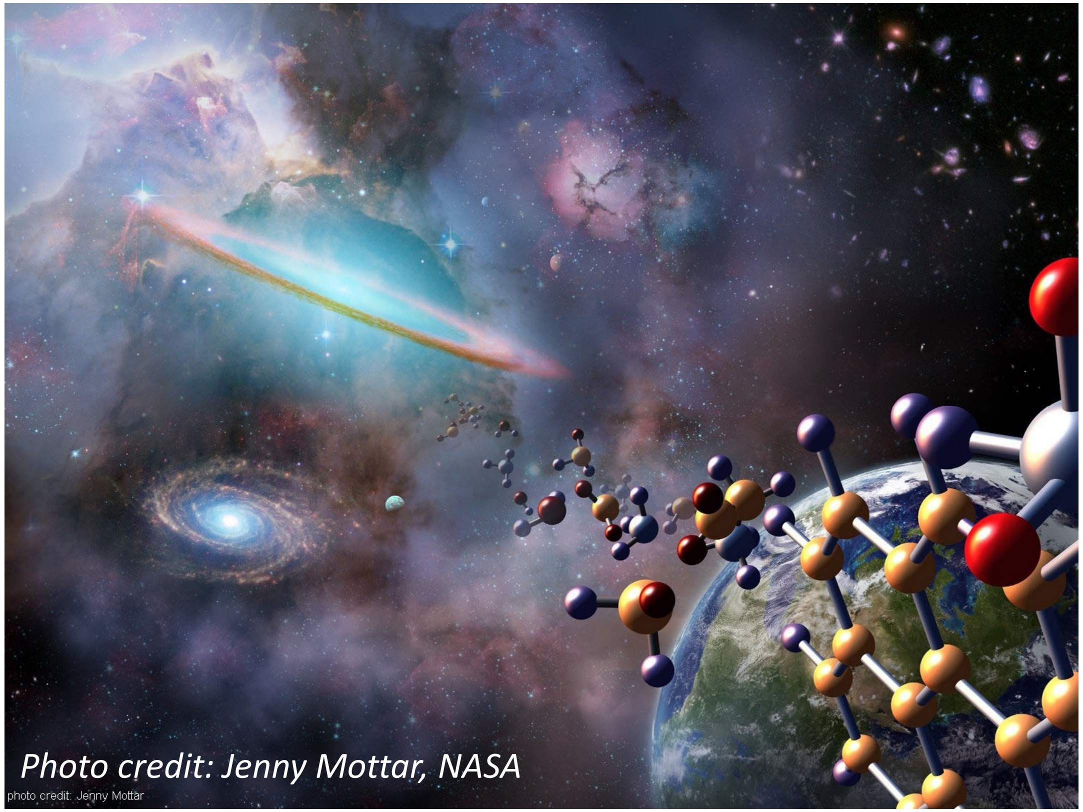
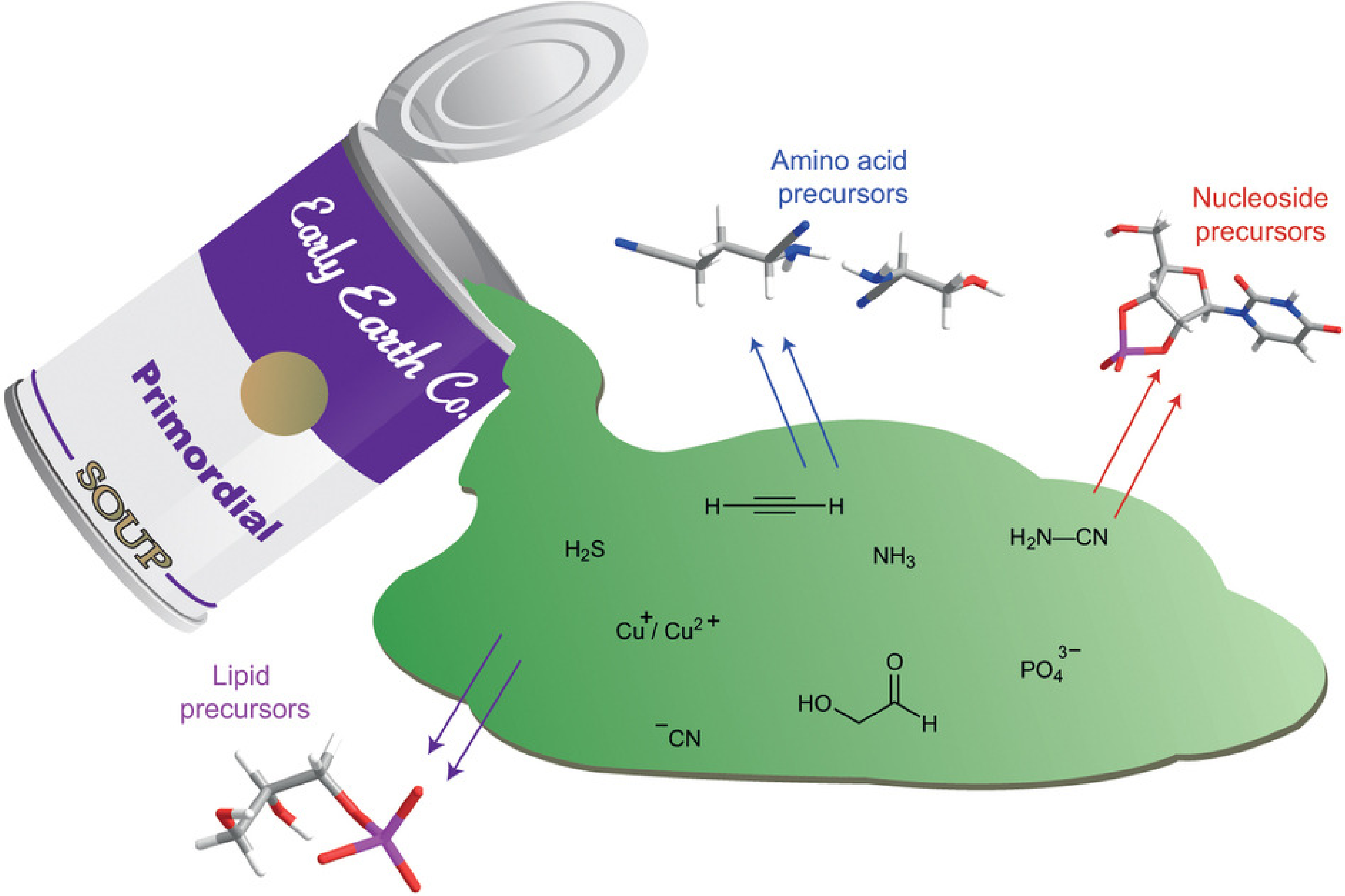


Photo credit: Jenny Mottar, NASA

L3 SoSe 2019 HD

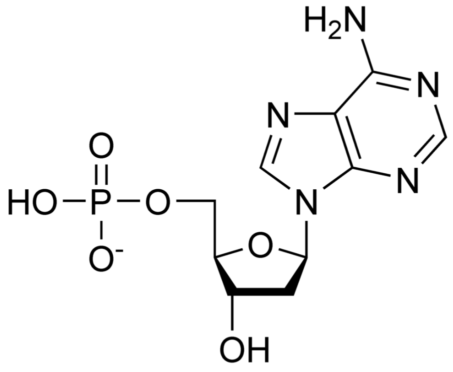
Zbigniew Pianowski

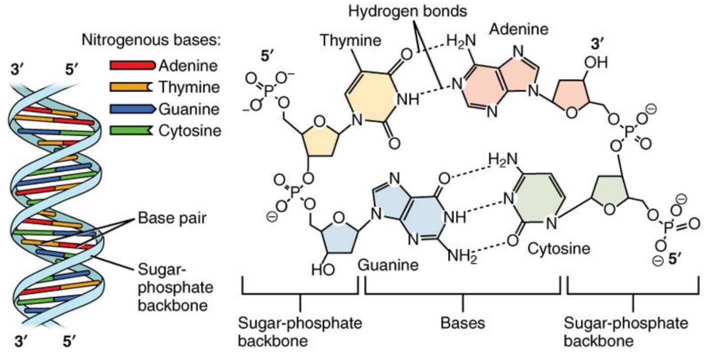
Basic classes of biomolecules

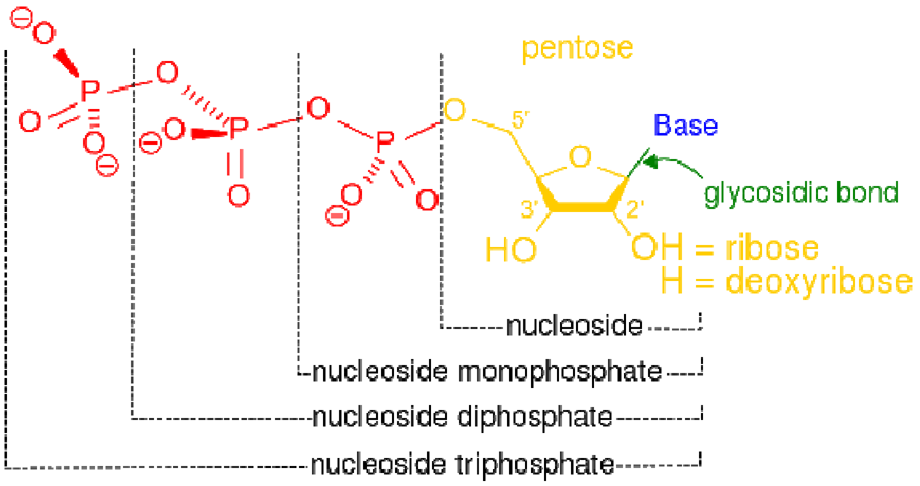


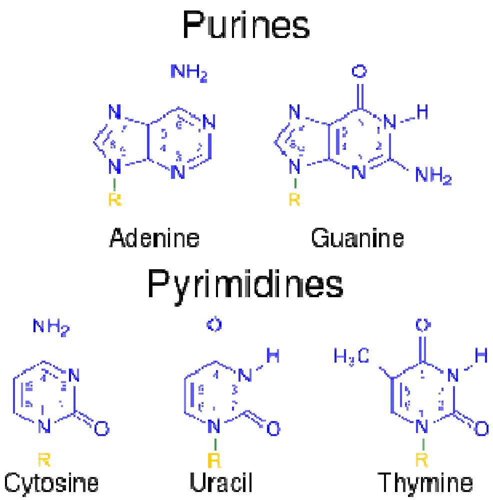
- Aminoacids
- Lipids
- Carbohydrates (sugars)
- Nucleotides
- Nucleosides (sugar+nucleotide)

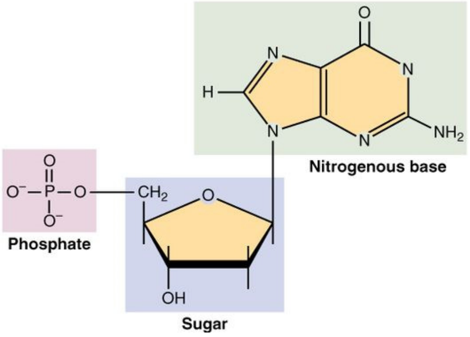
Nucleotides - components



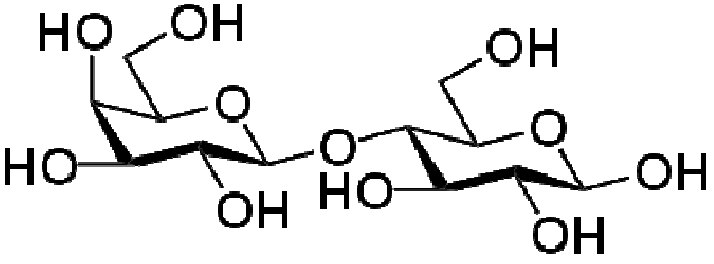


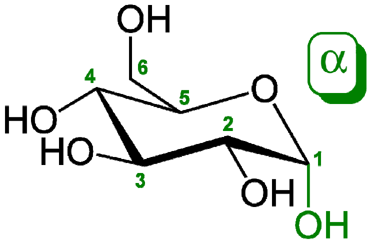


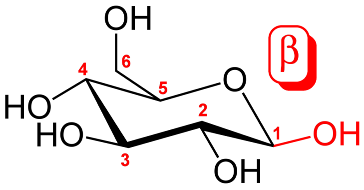




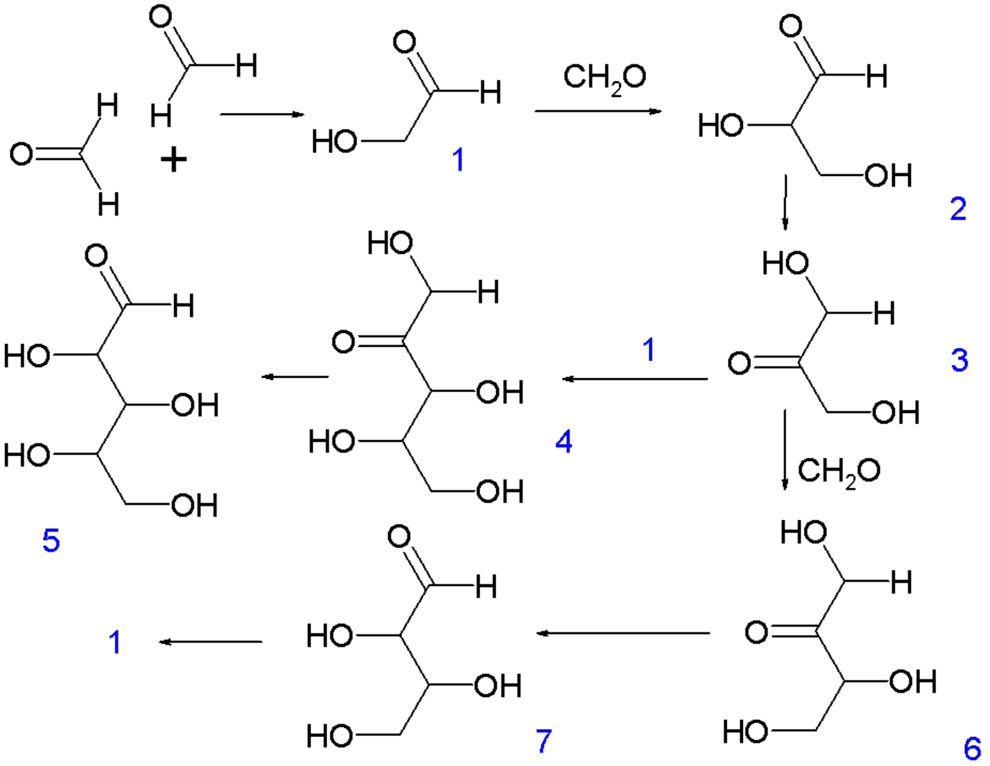
Carbohydrates







Formose reaction

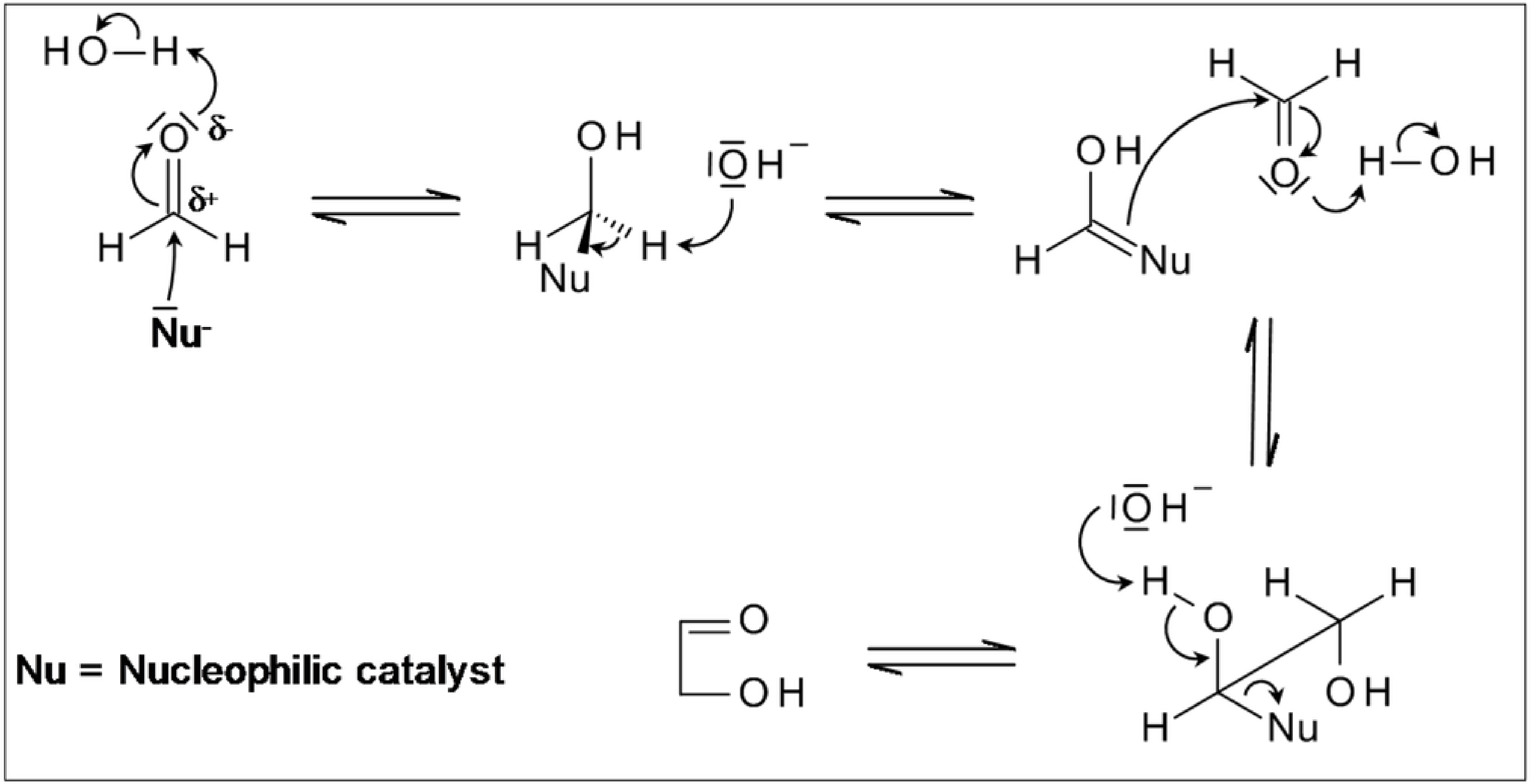


The reaction begins with two formaldehyde molecules condensing to make glycolaldehyde **1** which further reacts in an aldol reaction with another equivalent of formaldehyde to make glyceraldehyde **2**. An aldose-ketose isomerization of **2** forms dihydroxyacetone **3** which can react with **1** to form ribulose **4**, and through another isomerization ribose **5**. Molecule **3** also can react with formaldehyde to produce tetrulose **6** and then aldoltetrose **7**. Molecule **7** can split into **2** in a retro-aldol reaction.

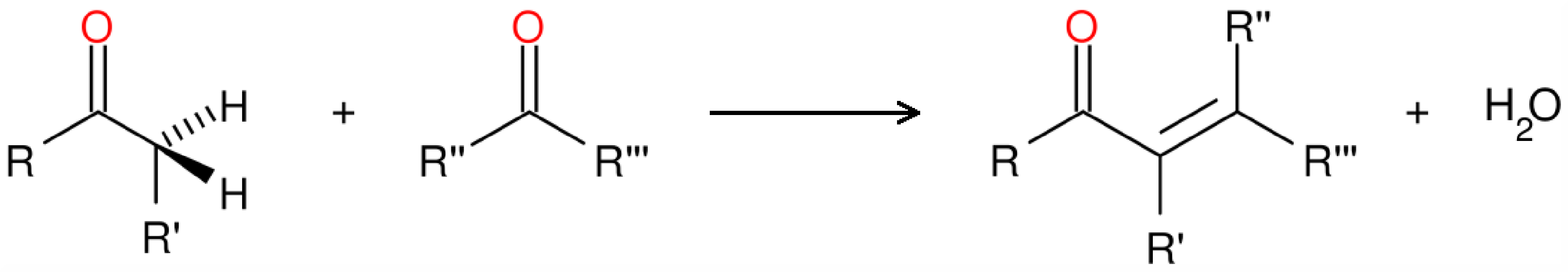
Alexander Butlerov (1828-1886)
St. Petersburg, Kazan, Russia

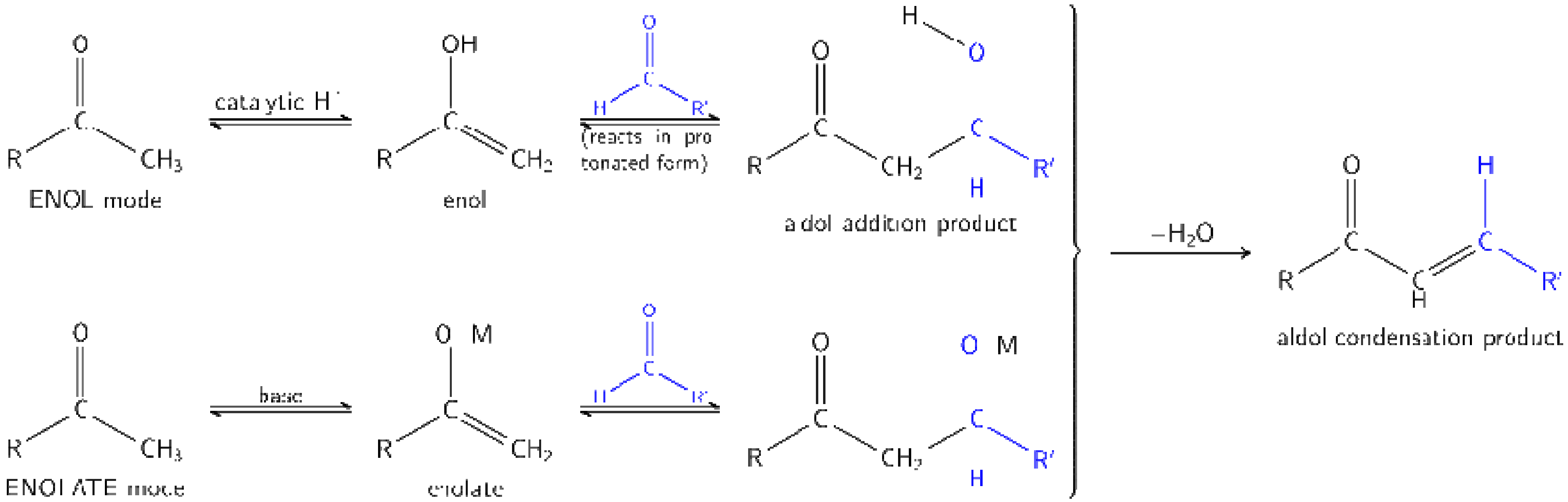
Ronald Breslow (1931-)
Columbia University, USA

Formaldehyde condensation



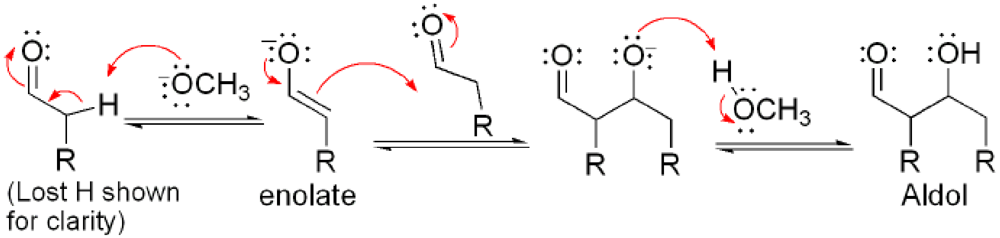
Aldol reaction



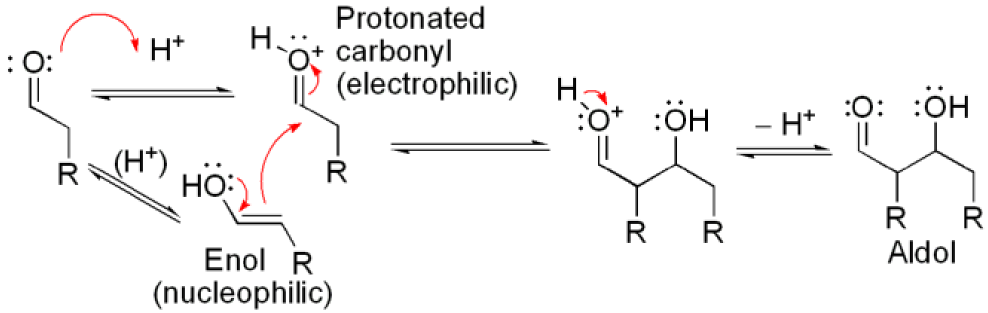


Aldol reaction

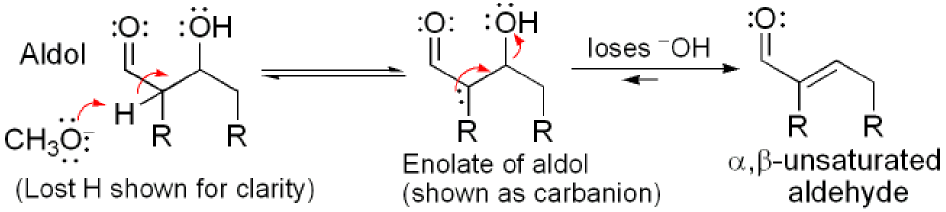
Base catalyzed aldol reaction (shown using $^{-}OCH_3$ as base)



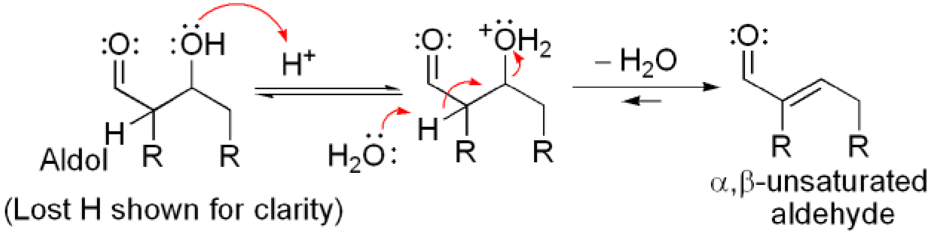
Acid catalyzed aldol reaction



Base catalyzed dehydration (sometimes written as a single step)

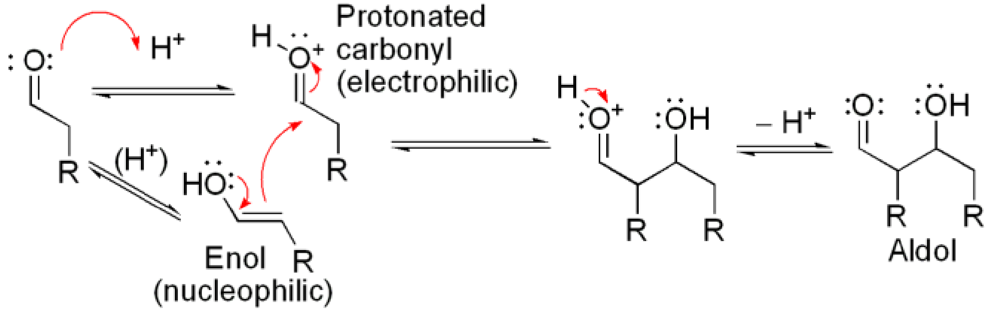


Acid catalyzed dehydration



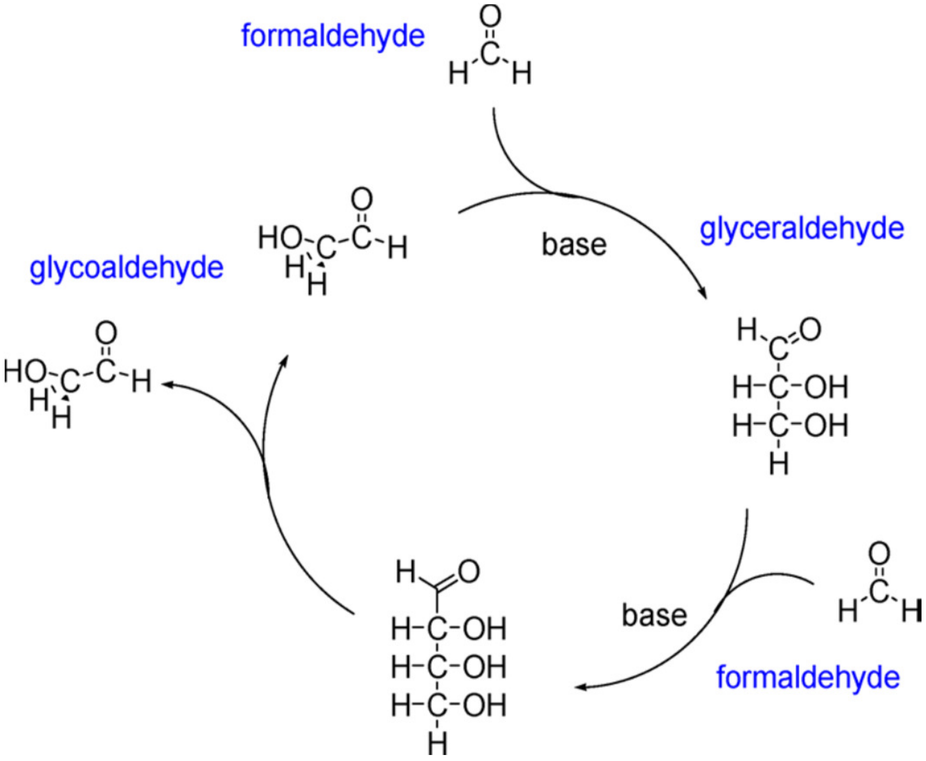
Reversible process
→ retro-aldol reaction

Acid catalyzed aldol reaction



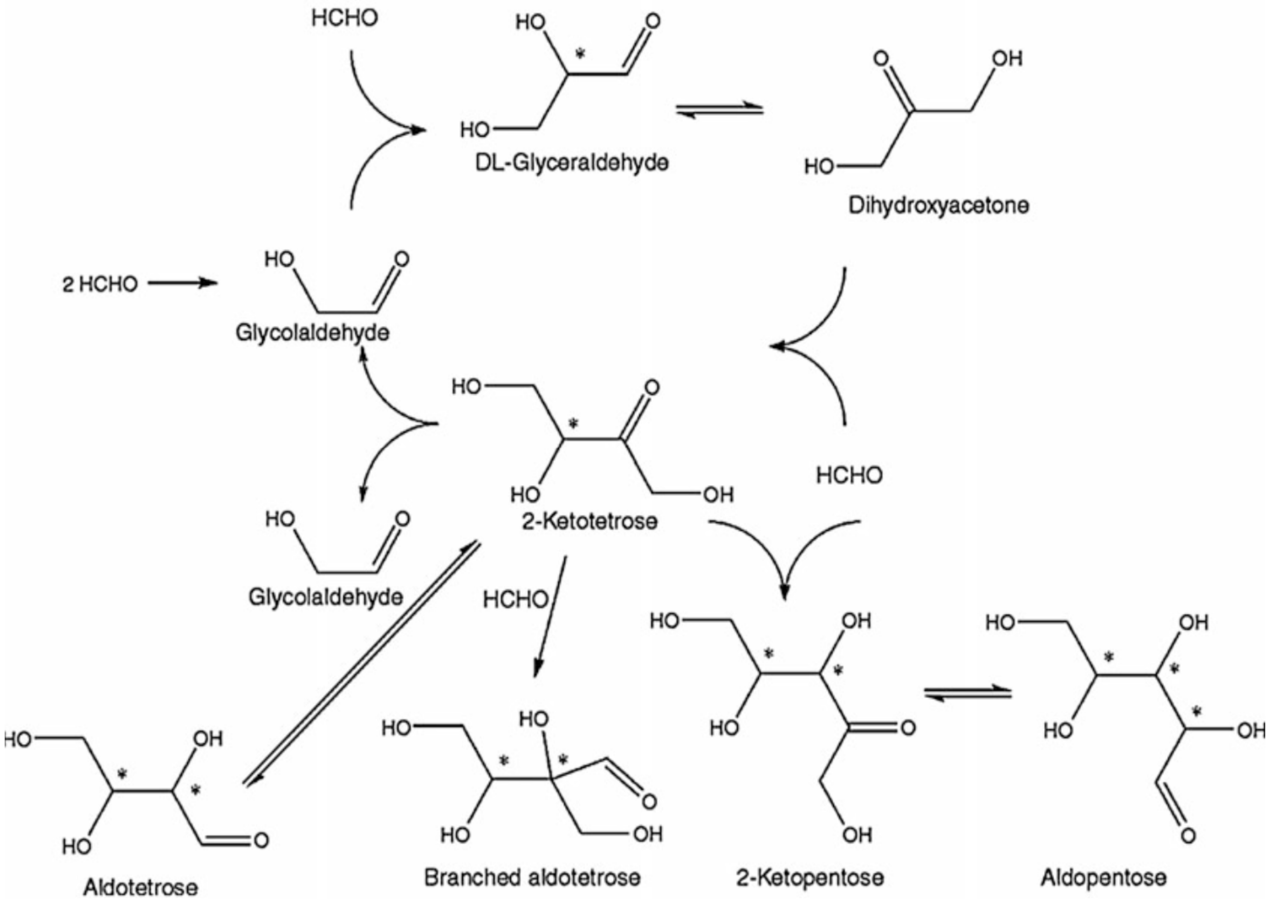
Formose reaction as an autocatalytic process

Formose reaction starts in concentrated alkaline aqueous solutions of formaldehyde
alkali are typically calcium, magnesium or lead



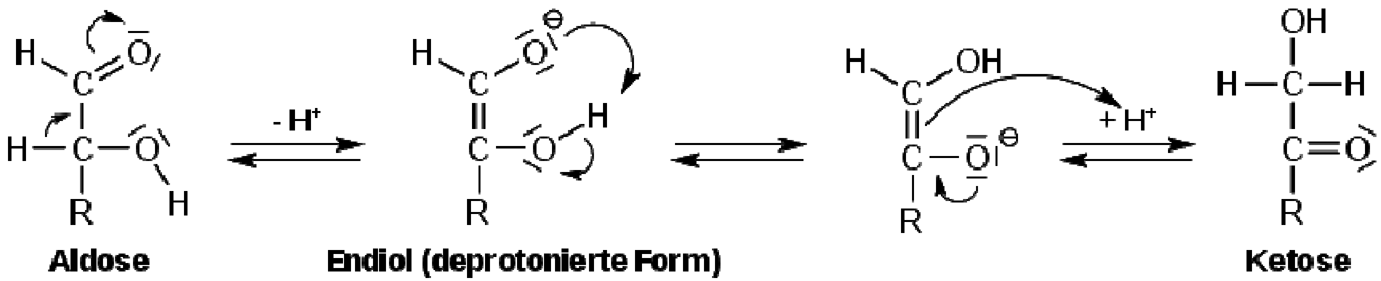
Detailed conditions for the formose reaction

Formose reaction starts in concentrated alkaline aqueous solutions of formaldehyde
alkali are typically calcium, magnesium or lead

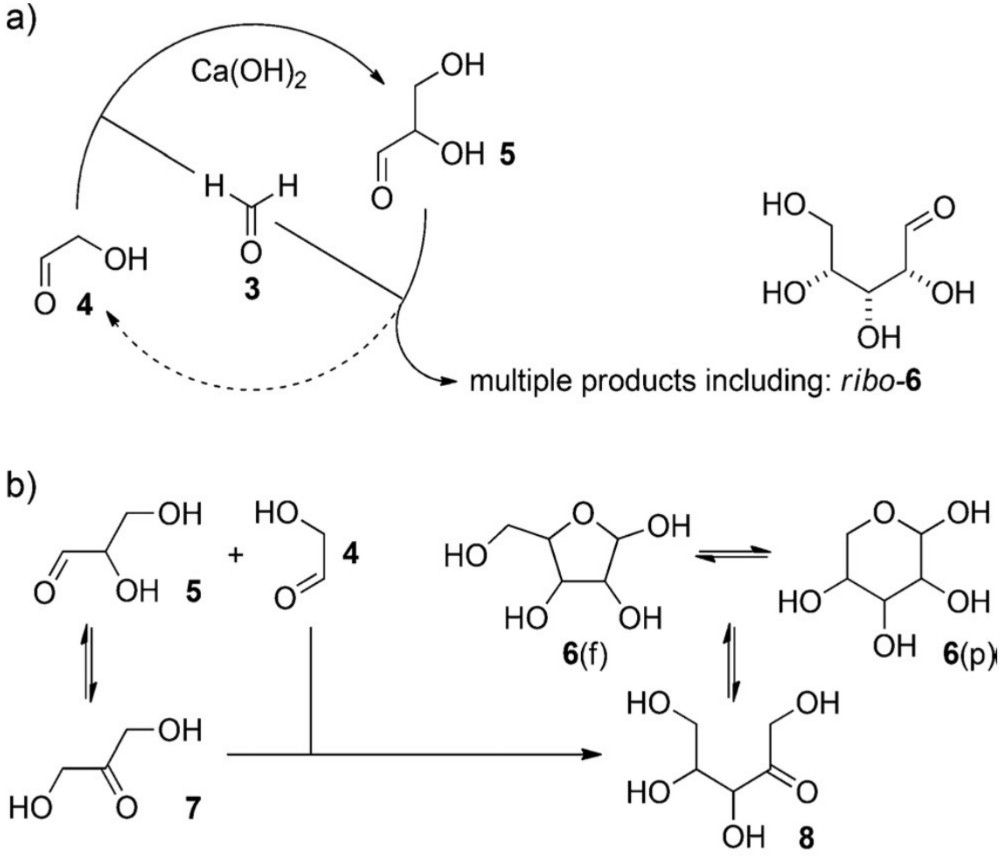


Aldose-ketose isomerization

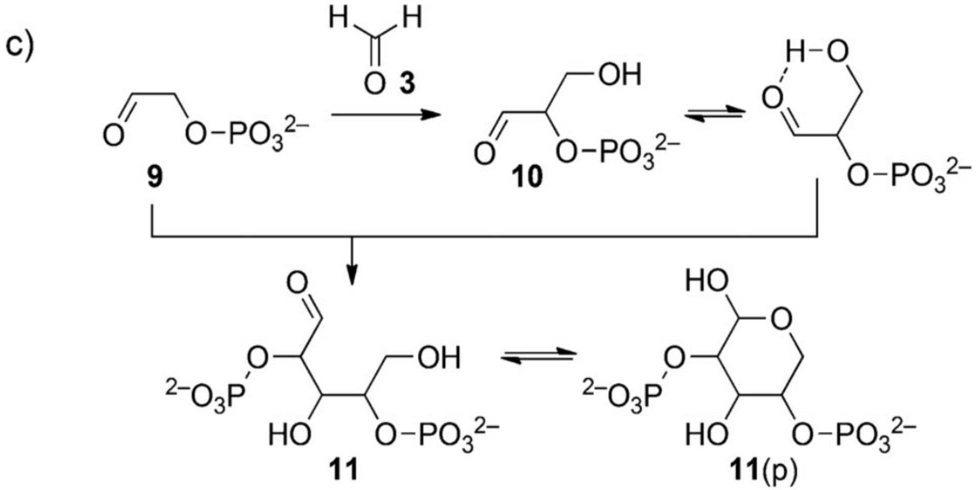
Lobry de Bruyn(–Alberda)-van Ekenstein transformation



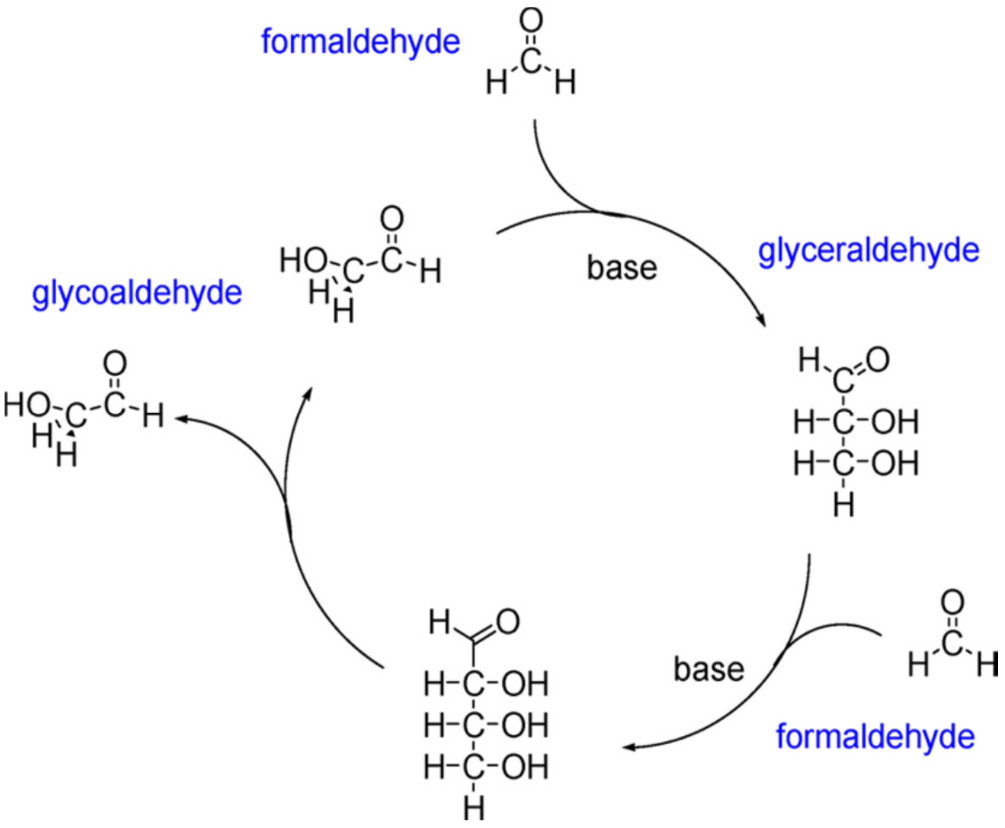
Formose reaction – substrate modifications

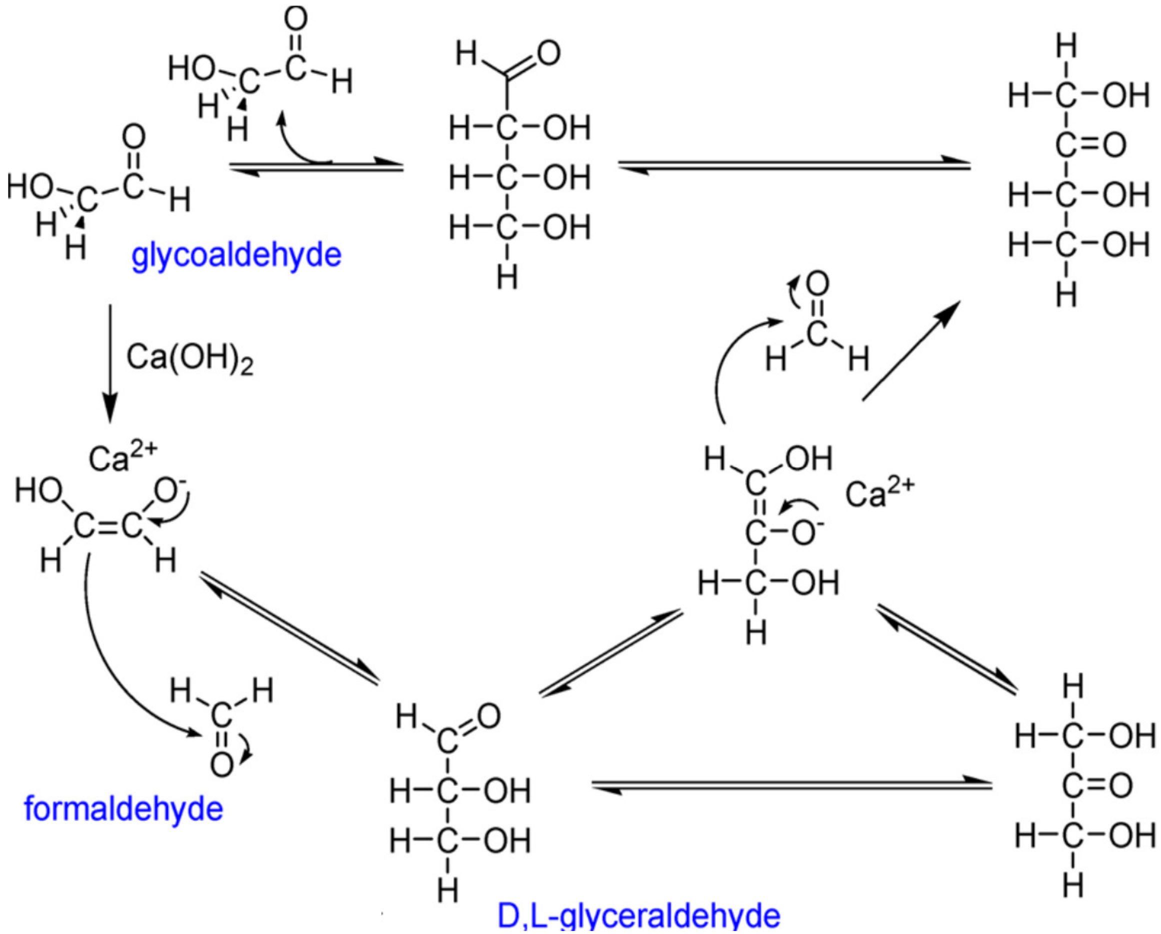


Formose reaction – substrate modifications



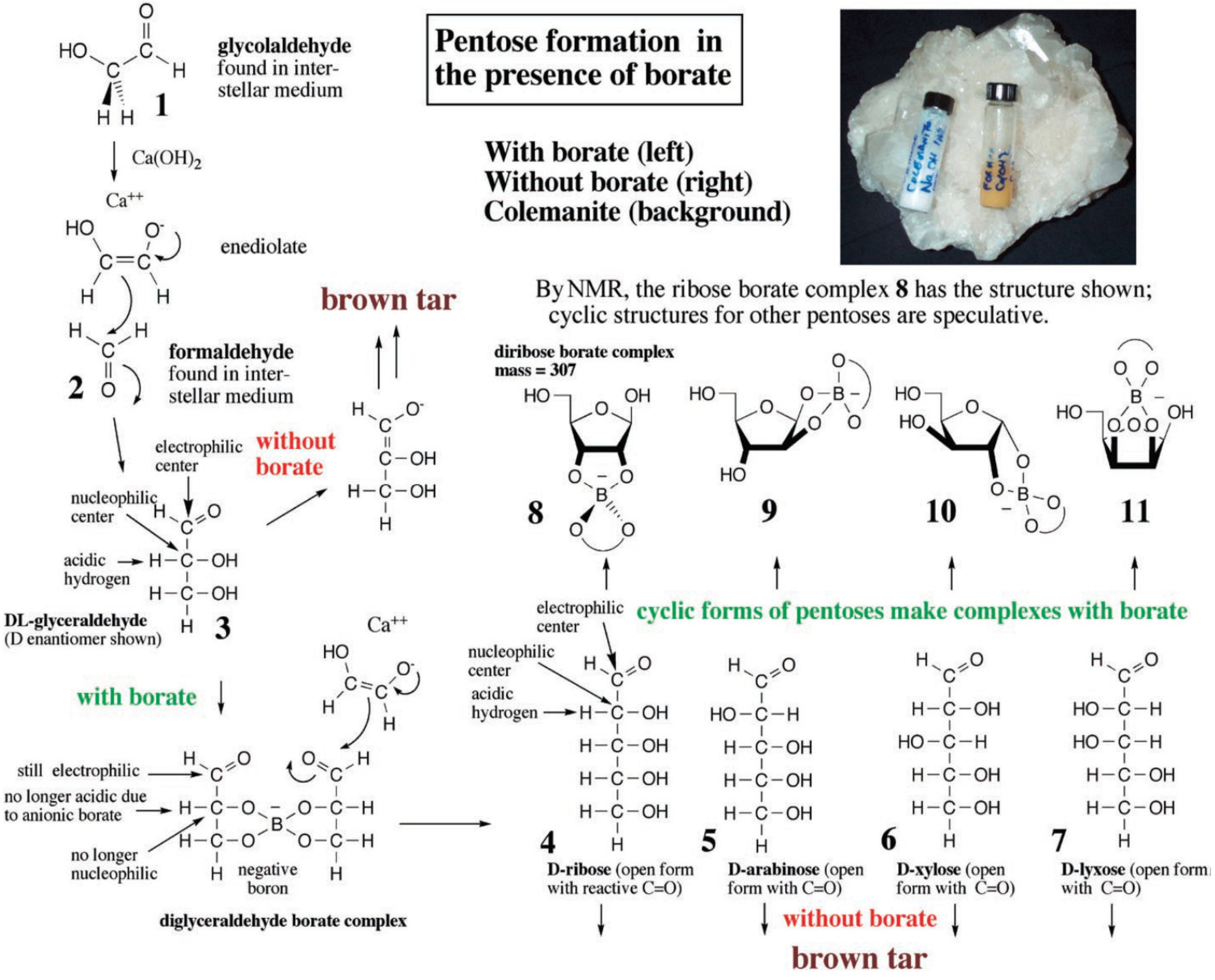
Formose reaction under standard basic catalysis





Escosura

Formose reaction in presence of borates



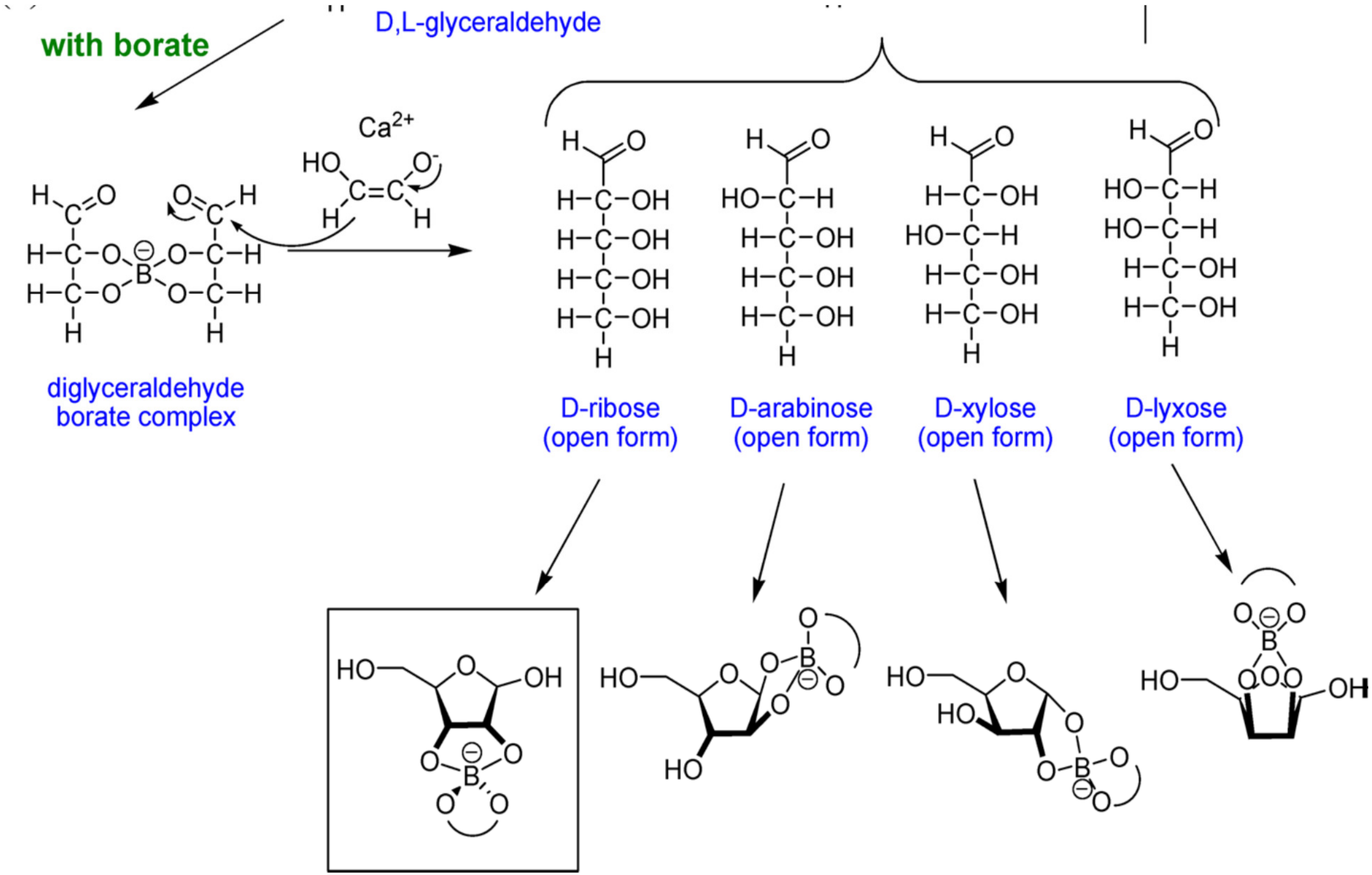
Pentose formation in the presence of borate

With borate (left)
Without borate (right)
Colemanite (background)

By NMR, the ribose borate complex **8** has the structure shown; cyclic structures for other pentoses are speculative.

A. Ricardo, M. A. Carrigan, A. N. Olcott, S. A. Benner *Science* **2004**, *303*, 196

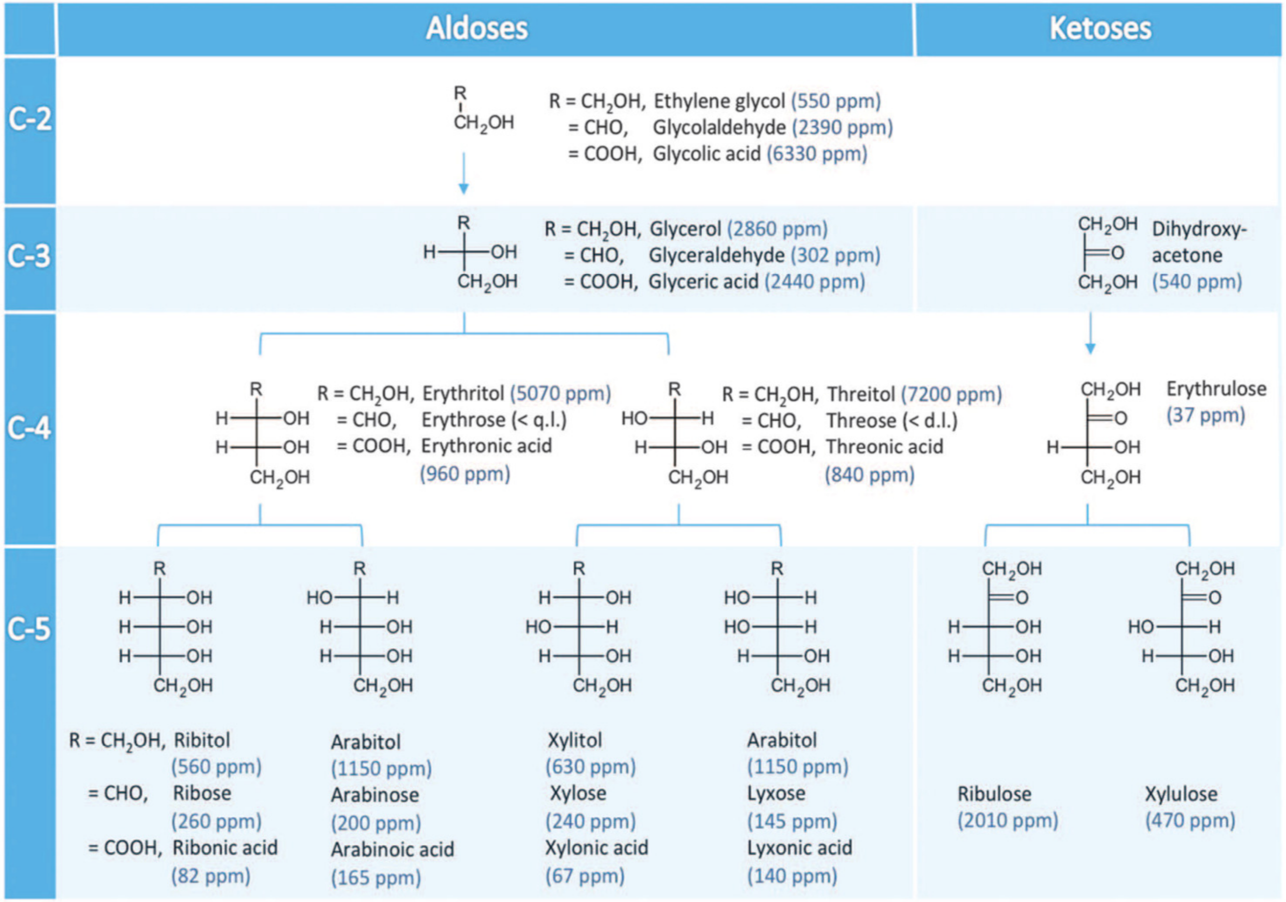
Formose reaction in presence of borates



Escosura

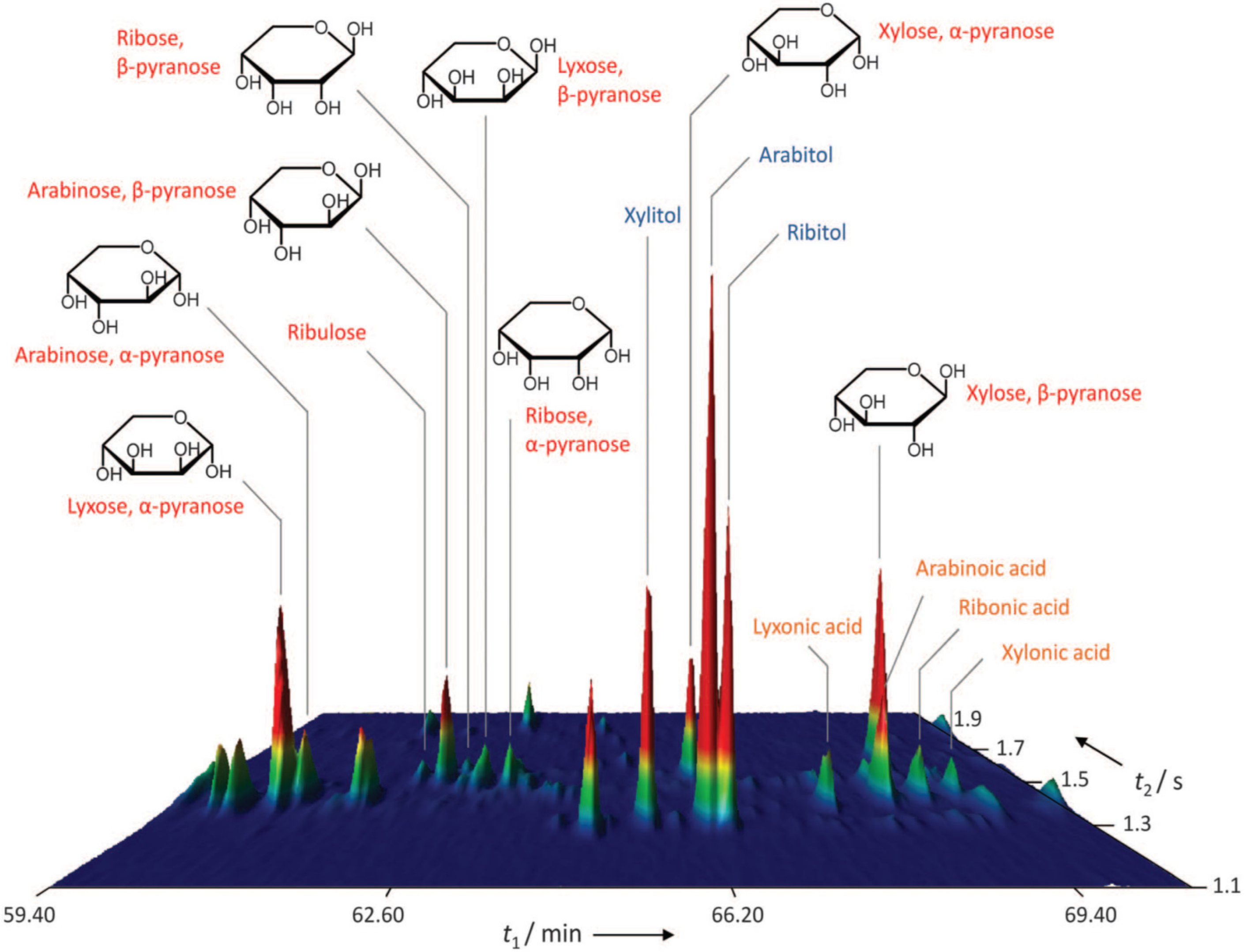
Interstellar origin of carbohydrates

Simulations of photo- and thermochemistry of interstellar ice (silicate/carbon grains surrounded by ice H_2O, CH_3OH and NH_3) exposed on UV light, low pressure (10^{-7} bar) and temperature (78K) delivers a mixture of sugars



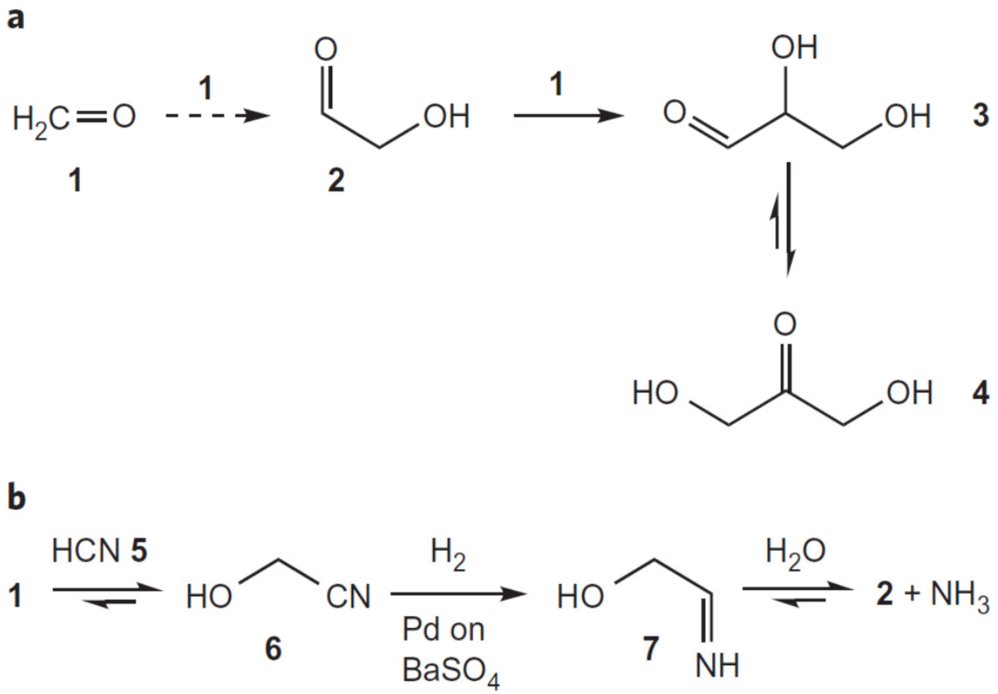
U. Meierhenrich, *et al. Science* **2016**, *352(6282)*, 208-212

Interstellar origin of carbohydrates



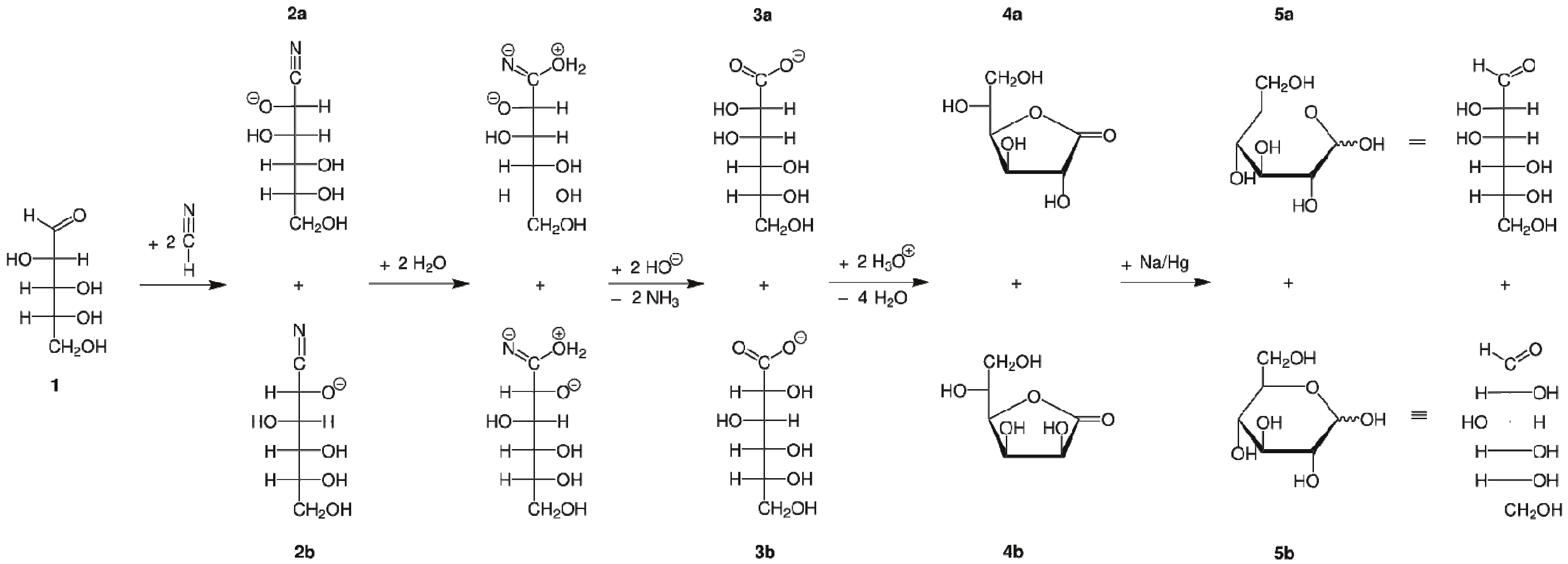
U. Meierhenrich, *et al. Science* **2016**, *352(6282)*, 208-212

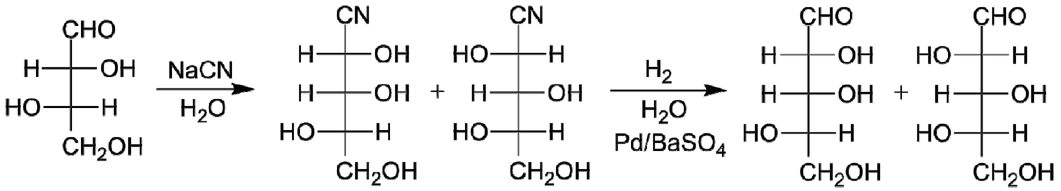
Prebiotically plausible alternatives to formose process



Homologation routes to simple sugars from formaldehyde **1**.
a, Direct homologation of formaldehyde **1** is problematic, because the first dimerization step (dashed) requires umpolung, and because the trimer is more stable as the ketose **4** than the aldose **3** under conditions where **3** can be formed from **1** and **2**. b, Kiliani–Fischer homologation of **1** in conventional synthetic chemistry involves favourable formation of the cyanohydrin **6** by reaction of **1** with hydrogen cyanide **5**, followed by the selective reduction of **6** using very specific conditions.

Kiliani-Fischer homologation

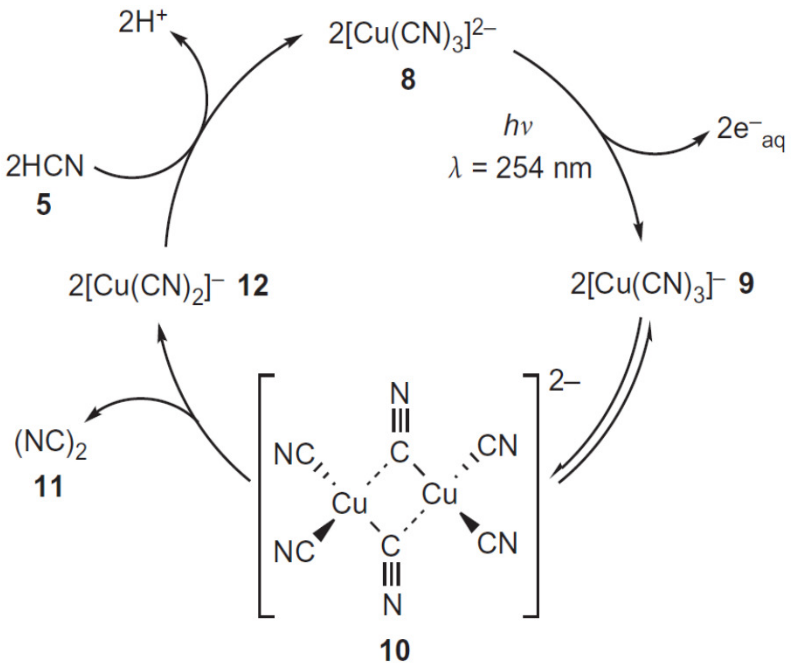


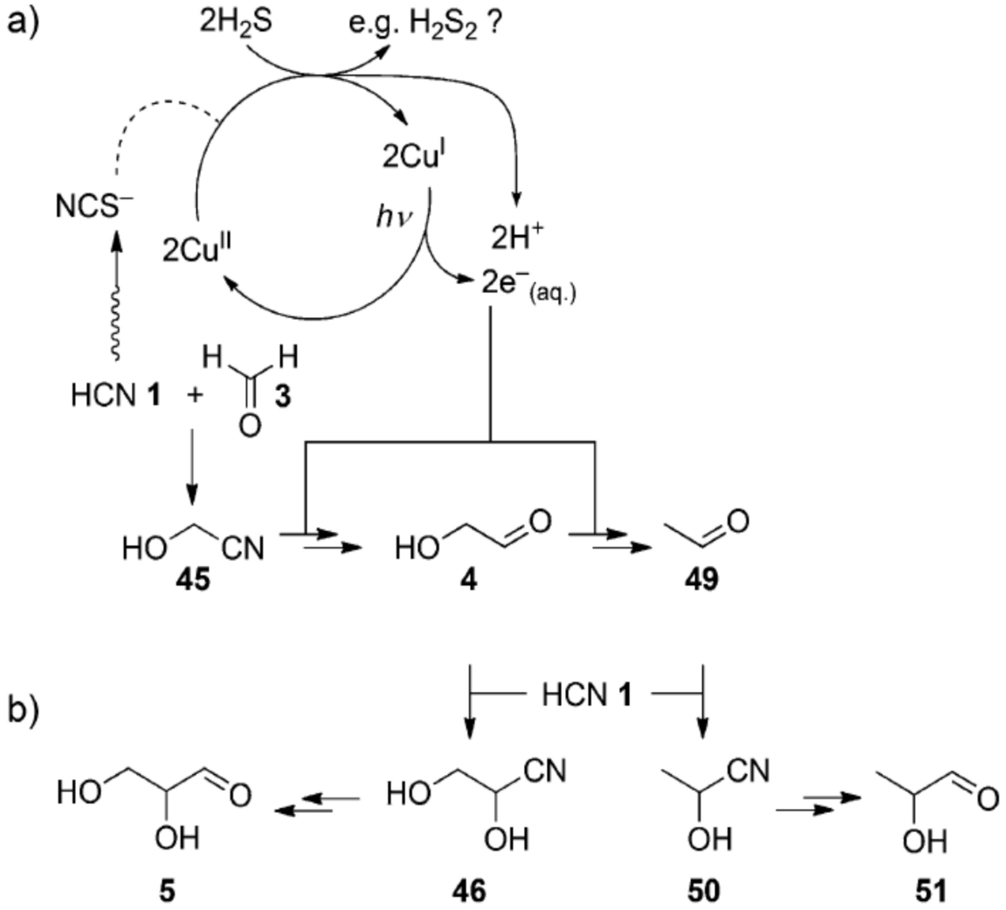


Cyanosulfidic chemistry for the Kiliani-Fischer homologation

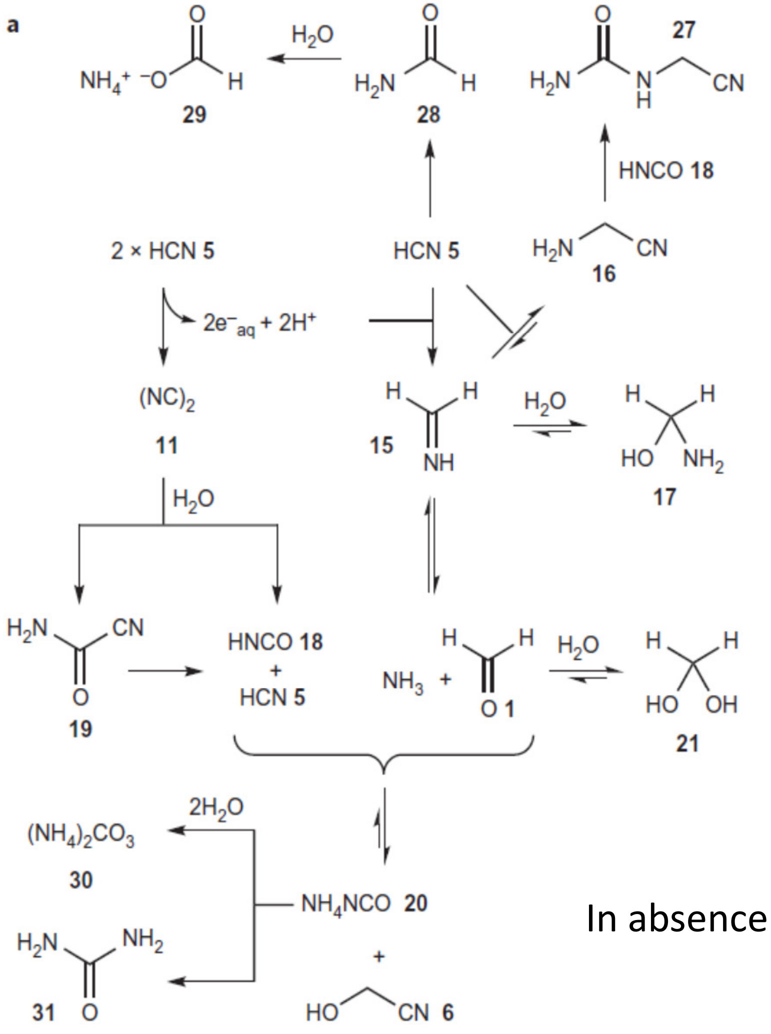
$Cu_2S + H_2O + 6CN^- \rightarrow 2[Cu(CN)_3]^{2-} + HS^- + OH^-$

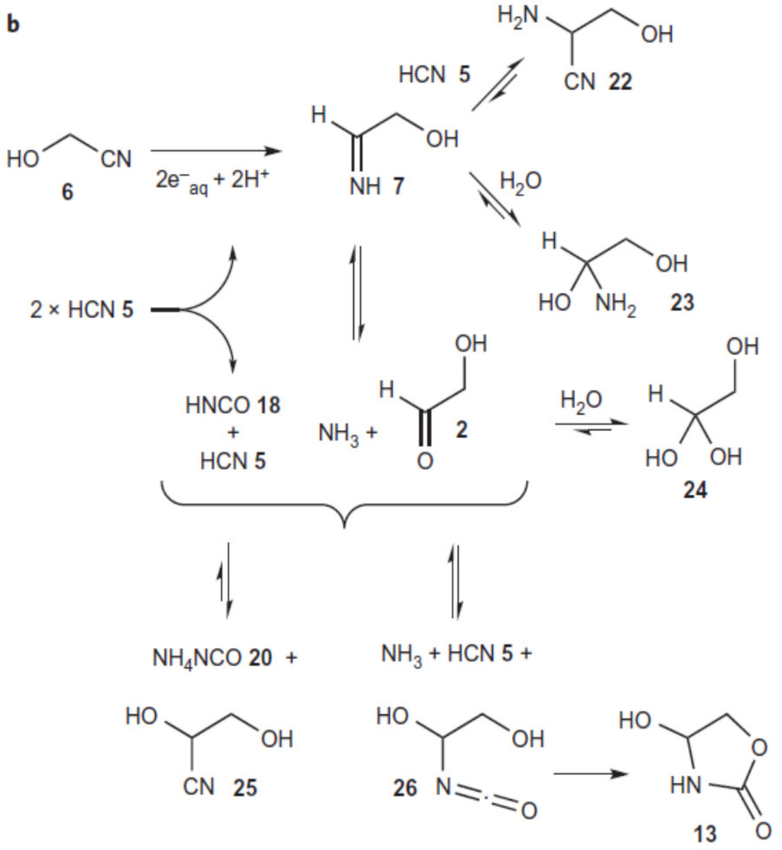
Photoredox cycle based on cyanocuprates may convert HCN into cyanogen





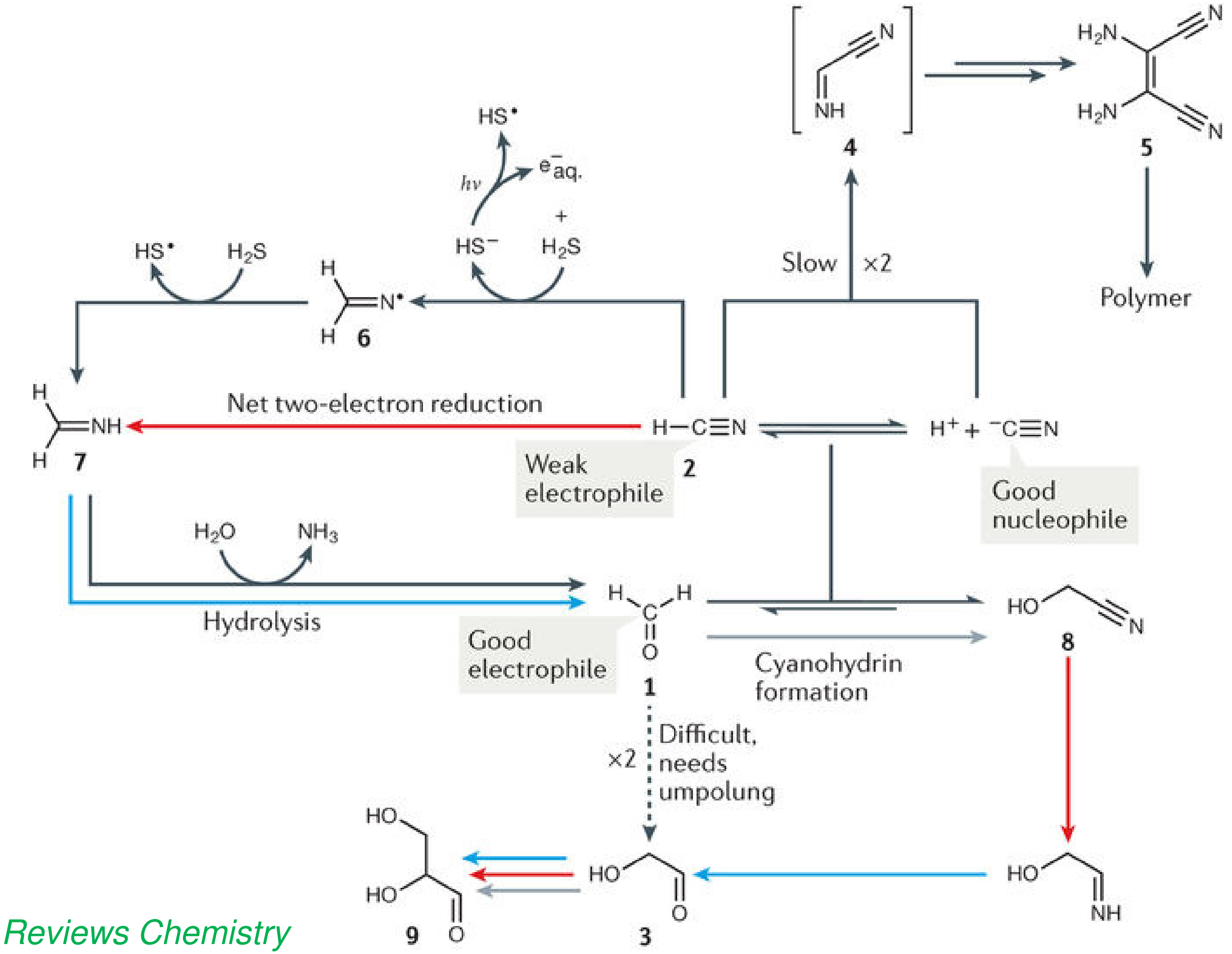
Cyanosulfidic chemistry for the Kiliani-Fischer homologation





In absence of HS^- glycolaldehyde **2** ends up mainly as a cyclic derivative **13**

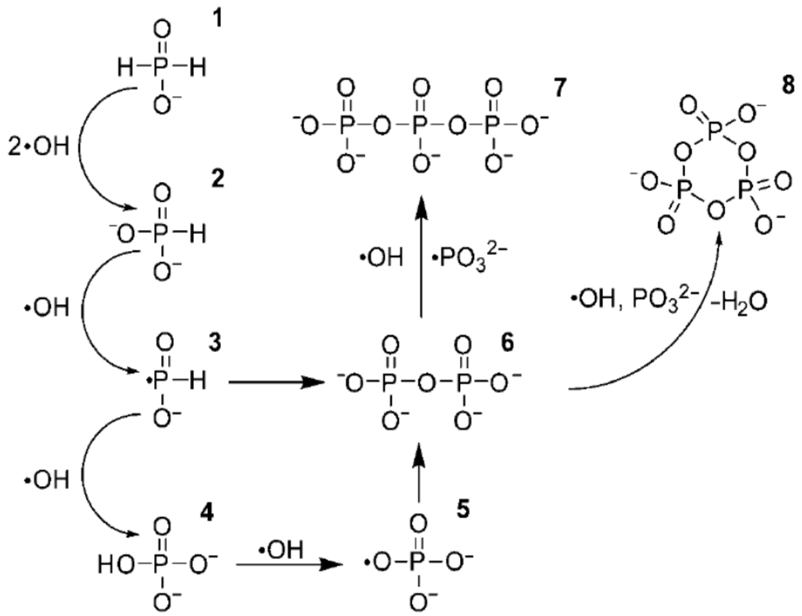
Cyanosulfidic chemistry for the Kiliani-Fischer homologation

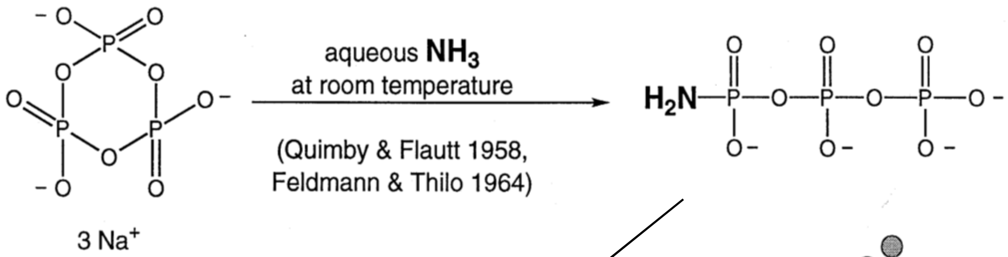


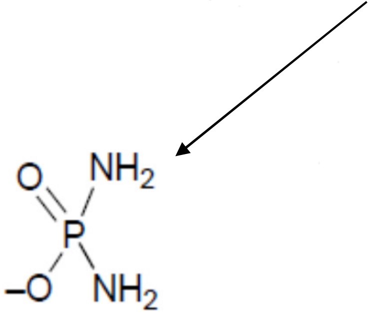
Nature Reviews | **Chemistry**

J. Sutherland, *Nature Reviews Chemistry*
2017, *1*, Article 0012, doi:10.1038/s41570-016-0012

Phosphorylation reagents







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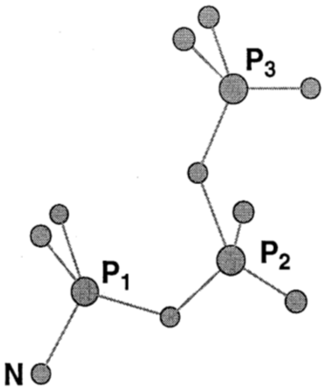
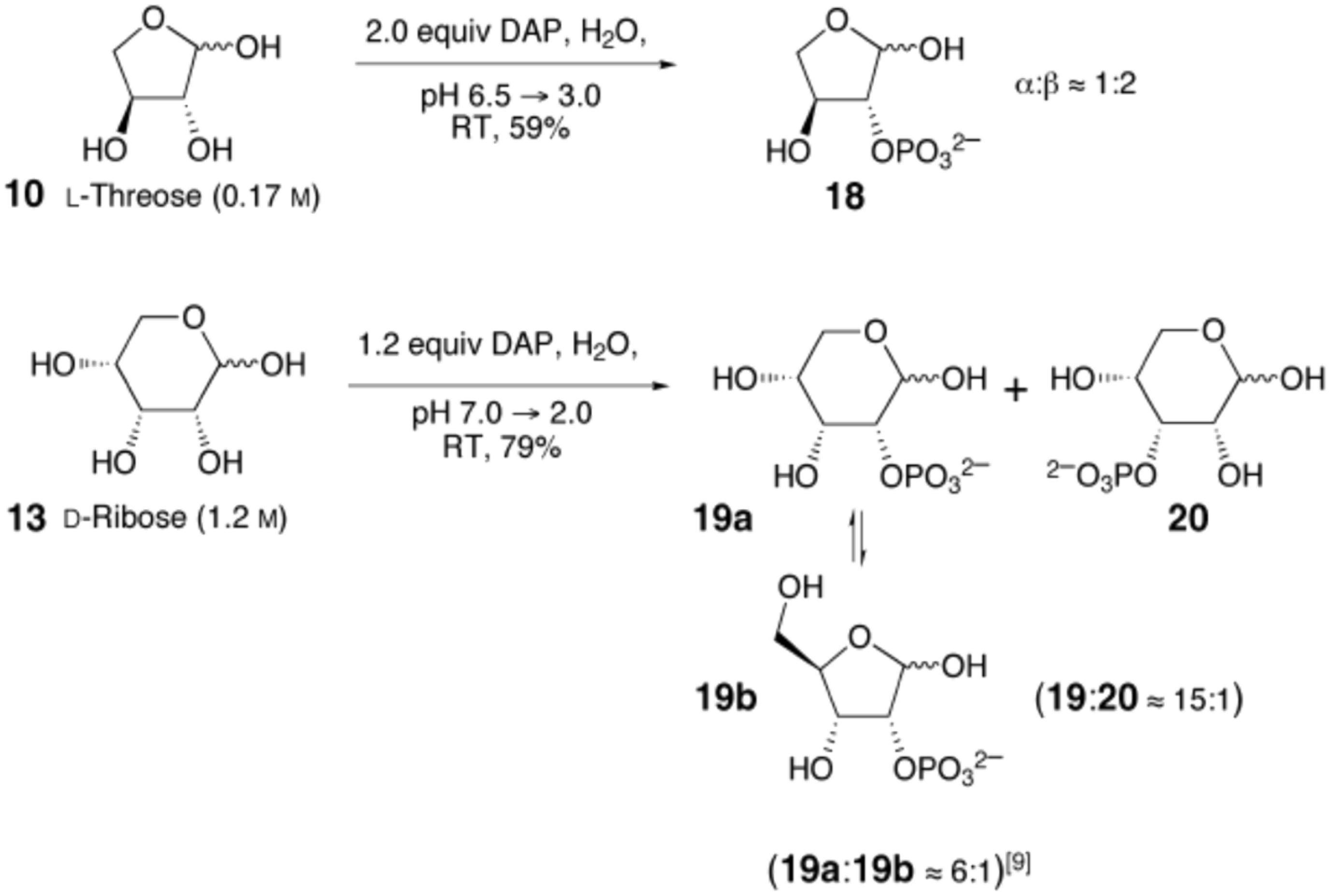
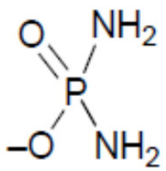


x-ray structure of $K_4(P_3O_9NH_2)$ (Hilmer, 1965)

M. A. Pasek, *et al. Angew. Chem. Int. Ed.* **2008**, *47*, 7918-7920 A. Eschenmoser, *et al. Orig. Life Evol. Biosph.* **1999**, *29*, 333-354

Phosphorylation reagents





DAP

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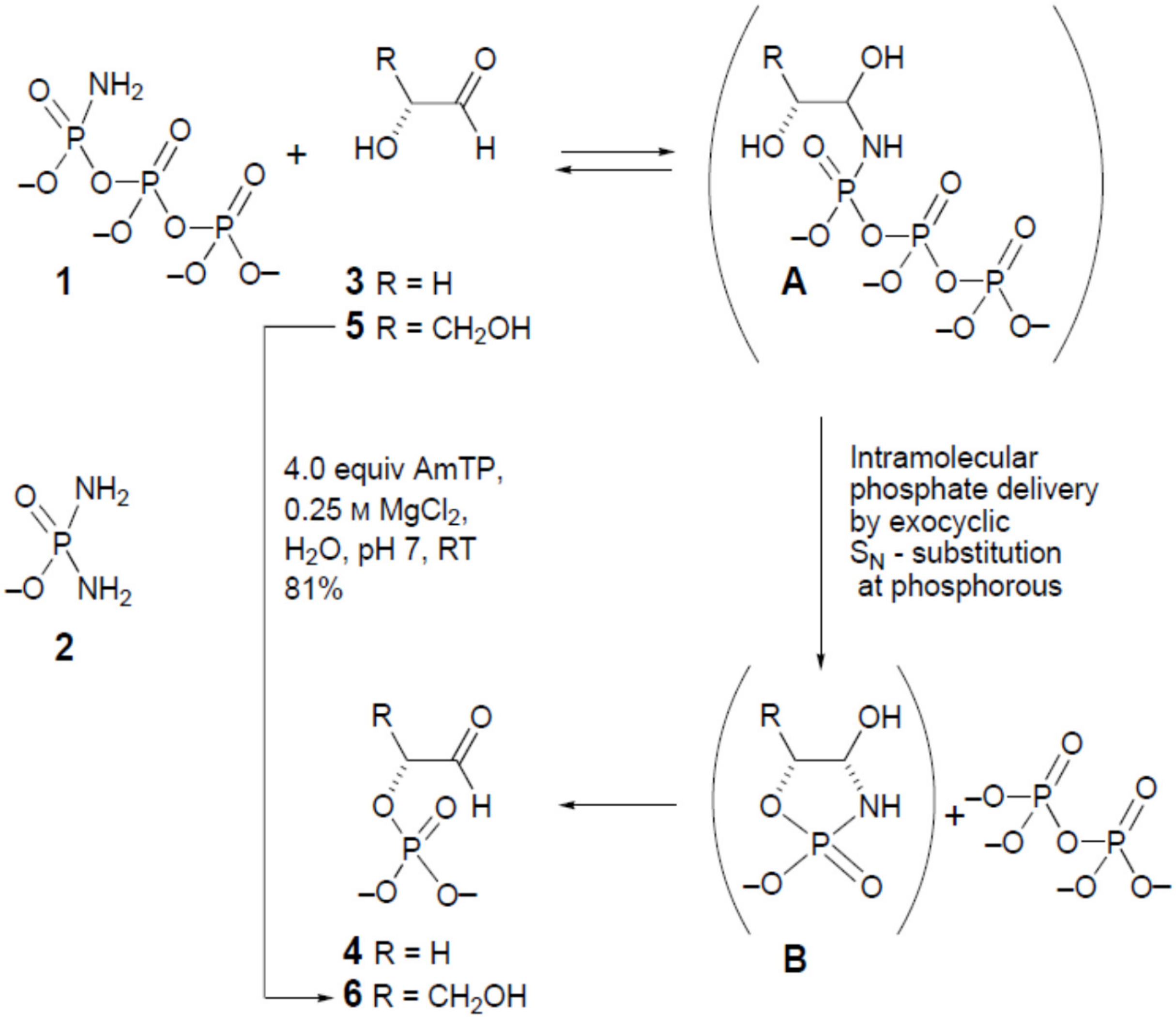
Phosphorylation of sugars



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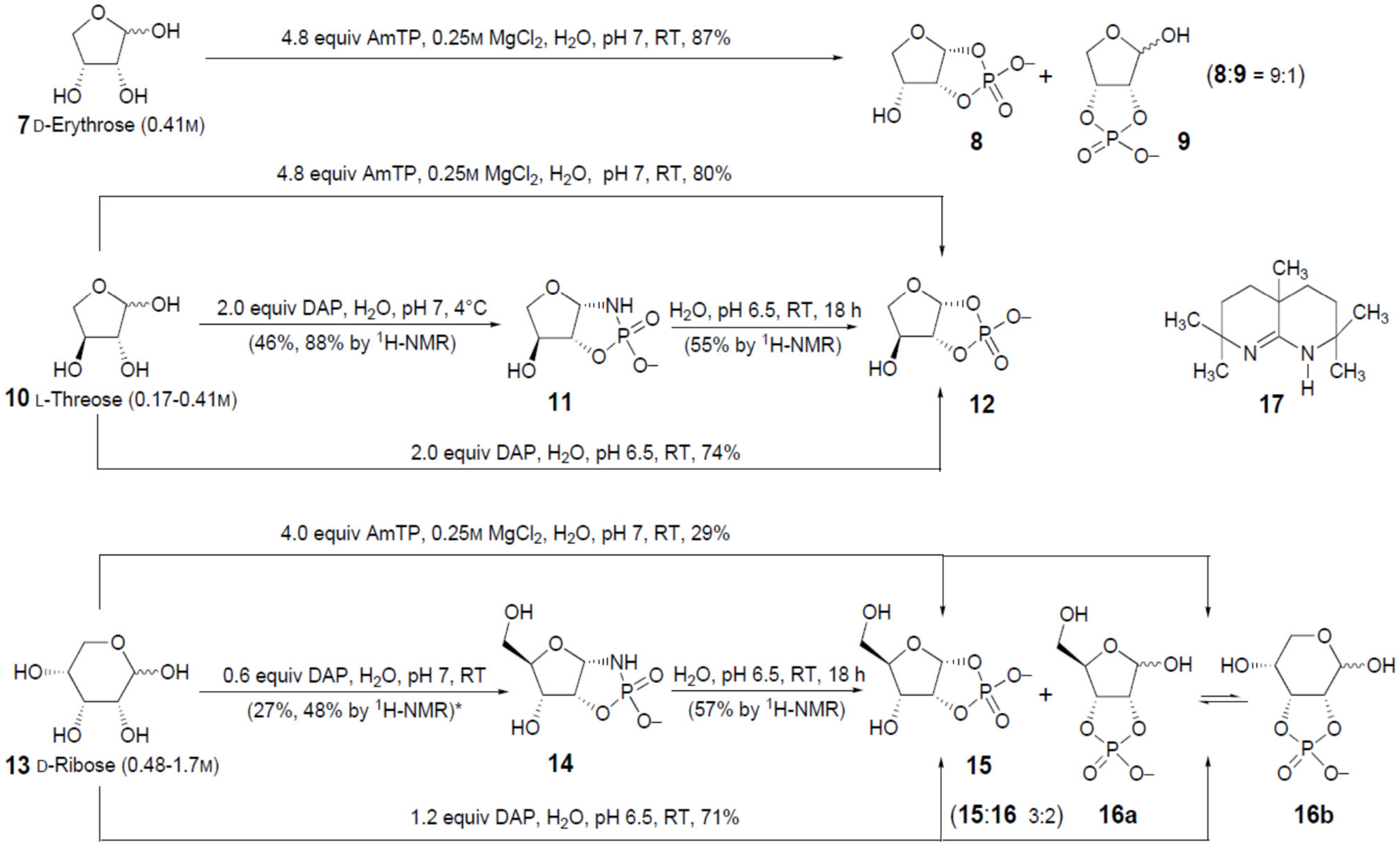
Phosphorylation of sugars



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Carbohydrates - summary

Formose reaction gives access to numerous C_2-C_5 and higher carbohydrates, but is difficult to direct towards particular outcome, and ultimately turns into polymeric tar if overcooked

In presence of borates, the formose reaction tends to deliver protected pentoses in high yields and stable form

Although formaldehyde is the simplest starting material, the reaction is autocatalytic in glycolaldehyde and without it long incubation period is required

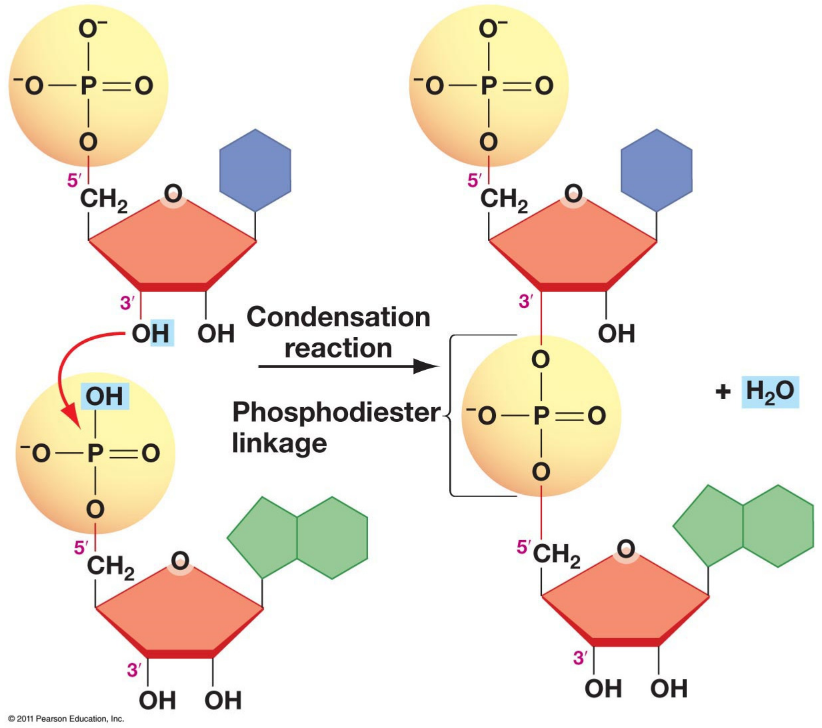
Carbohydrate synthesis can also occur under simulated extraterrestrial conditions – by UV-light irradiation of cometary ice

Alternative prebiotic synthesis of simple carbohydrates involves Kiliani-Fischer homologation process based on HCN in presence of copper ions and hydrosulfides – all accessible by the meteorite-derived cyanide-metal chemistry

The same type of chemistry can also deliver a set of reactive intermediates like cyanogen, acetylene, ammonia, and activated forms of phosphate – the latest can derivatize sugars and, after redox processes, deliver numerous building blocks present in currently known metabolic cycles

Nucleotide polymerization

Regioselective formation of 3’-5’ phosphodiester bonds between nucleotides



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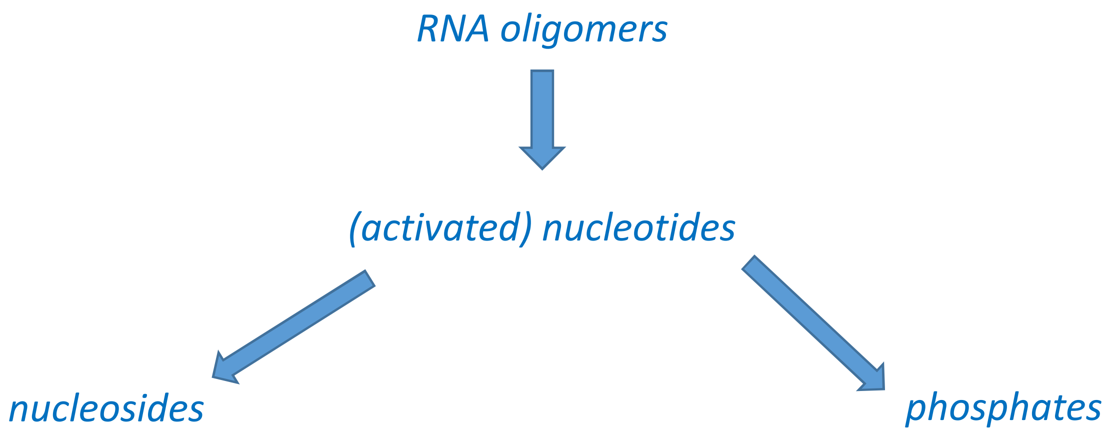
RNA oligomers – prebiotic disconnections



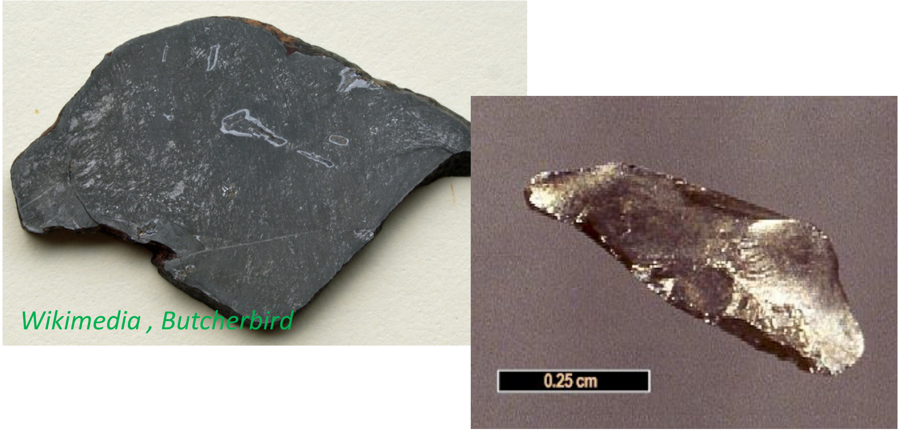
RNA oligomers

(activated) nucleotides

nucleosides

phosphates

Phosphates



Wikimedia , Butcherbird

Schreibersite $(Fe,Ni)_3P$, common in iron-nickel meteorites

HCN – burning of carbon-rich meteorites into redox-neutral atmosphere (N_2 and H_2O)

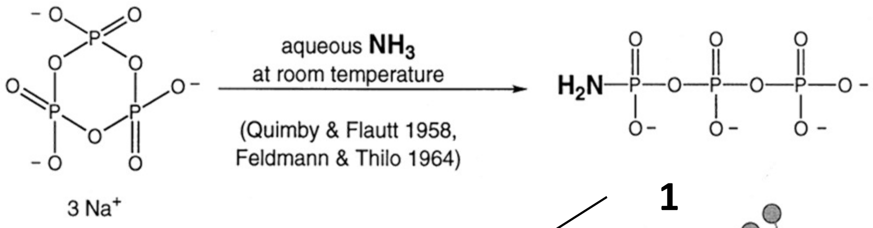
$(Fe,Ni)_3P + HCl_{aq} \rightarrow H_2PO_3^-$

$Fe^{2+} + PO_4^{3-}$ (pH ~ 7) $\rightarrow Fe_3(PO_4)_2$ (**vivianite**)

vivianite

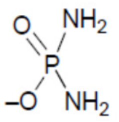
Wikimedia , Carles Millan

$Fe_3(PO_4)_2 + 18CN^-_{aq} \rightarrow 2PO_4^{3-} + 3[Fe(CN)_6]^{4-}$



1

M. A. Pasek, *et al. Angew. Chem. Int. Ed.* **2008**, *47*, 7918-7920



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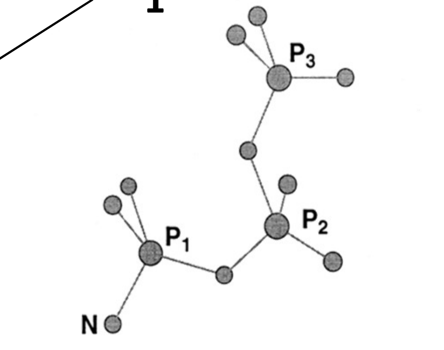

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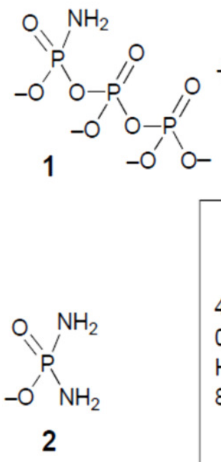
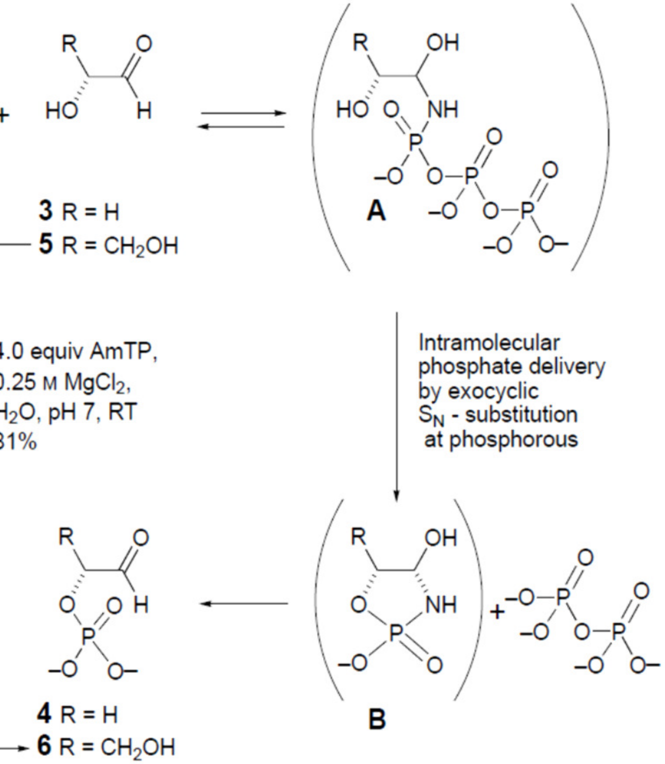
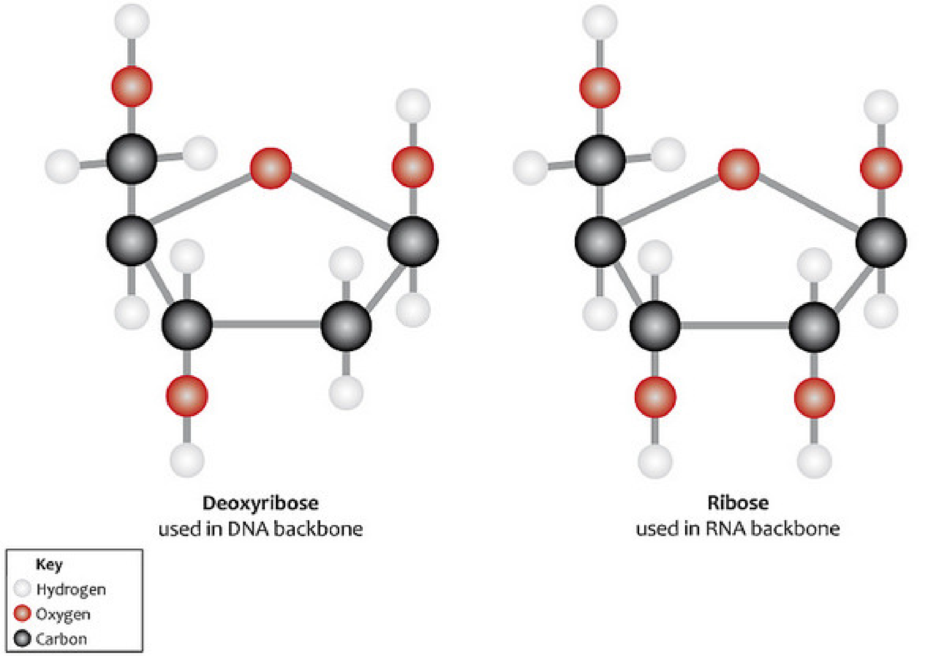
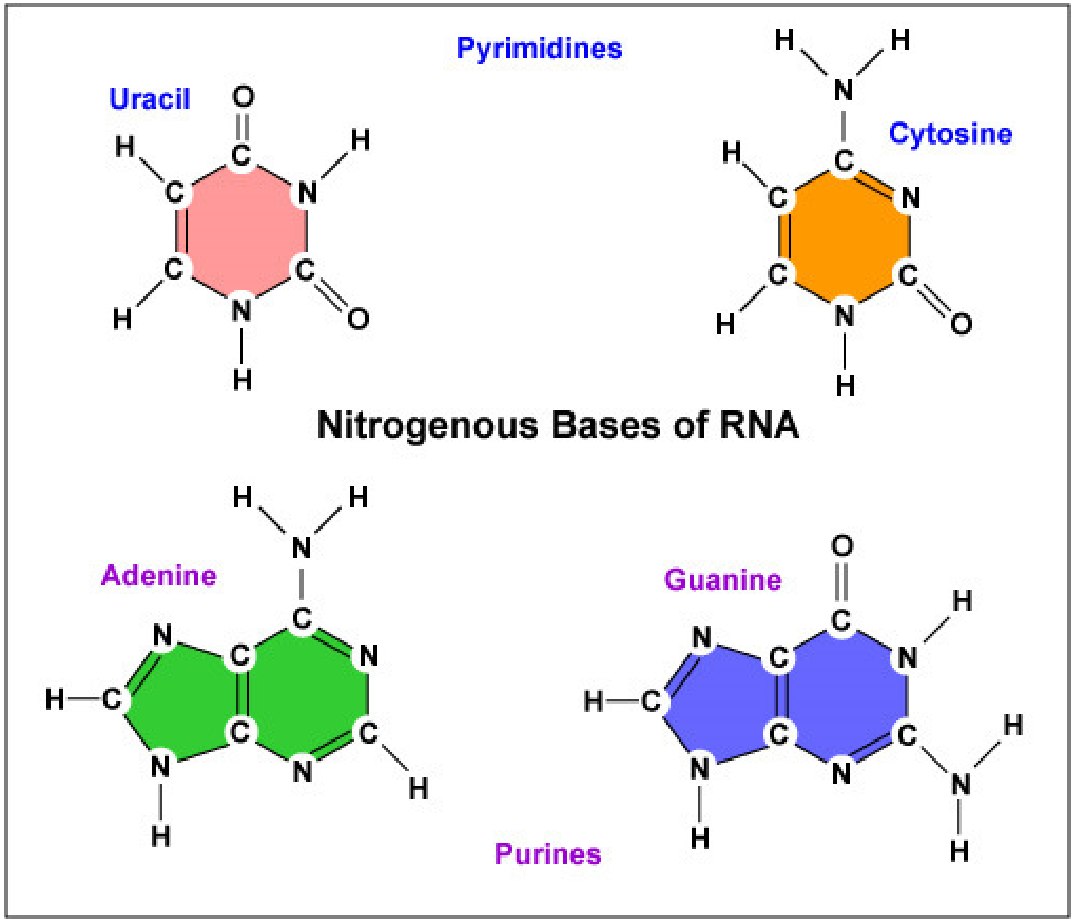




A. Eschenmoser, *et al. Orig. Life Evol. Biosph.* **1999**, *29*, 333-354

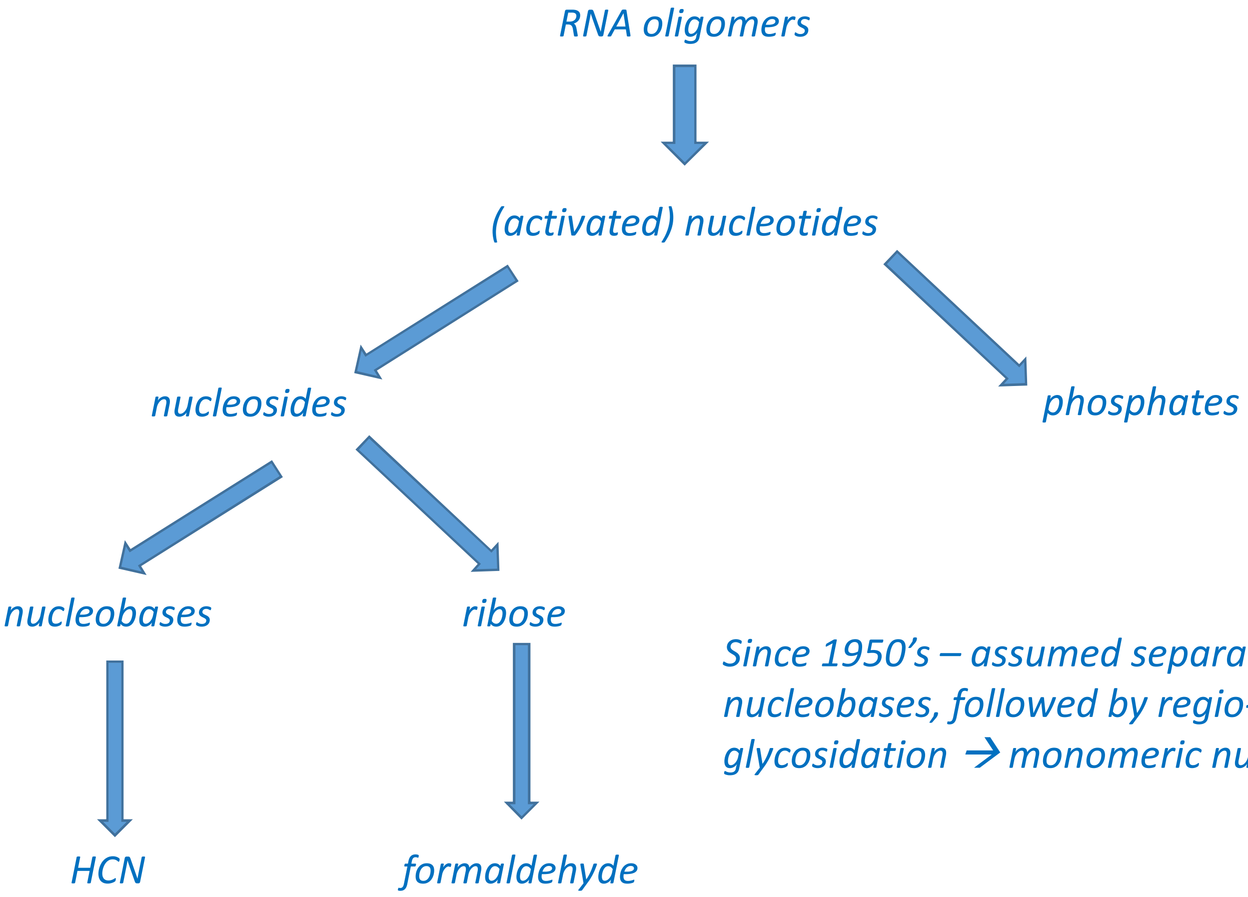
Nucleosides - nucleobases + sugars





Since 1950's – assumed separate synthesis of sugars and nucleobases, followed by regio- and diastereoselective glycosidation → monomeric nucleotides

RNA oligomers – prebiotic disconnections



RNA oligomers

(activated) nucleotides

nucleosides

phosphates

nucleobases

ribose

HCN

formaldehyde

Since 1950's – assumed separate synthesis of sugars and nucleobases, followed by regio- and diastereoselective glycosidation → monomeric nucleotides

Literature sources

J. Oro, *Biochem. Biophys. Res. Commun.* **1960**, *2*, 407–412.

J. D. Sutherland, *Cold Spring Harbor Perspect. Biol.* **2010**, 2, a005439.

M. W. Powner, B. Gerland, J. D. Sutherland, *Nature* **2009**, *459*, 239–242

J. D. Sutherland, *Angew. Chem. Int. Ed.* **2016**, *55*, 104-121.

B. H. Patel, C. Percivalle, D. J. Ritson, C. D. Duffy, J. D. Sutherland, *Nat. Chem*. **2015**, *7*, 301–307.

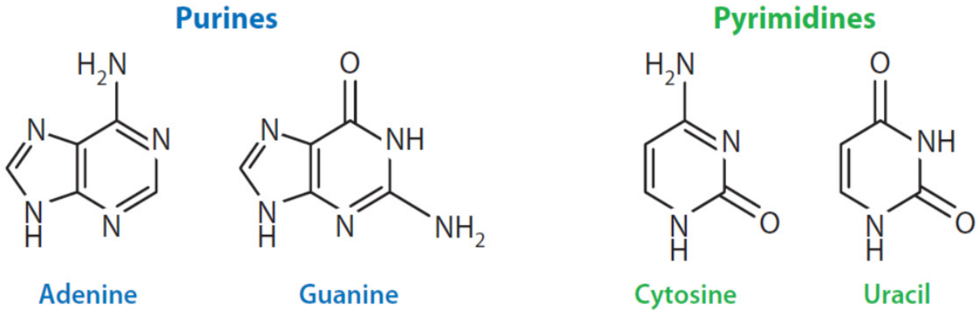
J. D. Sutherland, *et al. Nat. Chem*. **2013**, *5*, 383–389.

L. E. Orgel, *Crit. Rev. Biochem. Mol. Biol.* **2004**, *39*, 99-123.

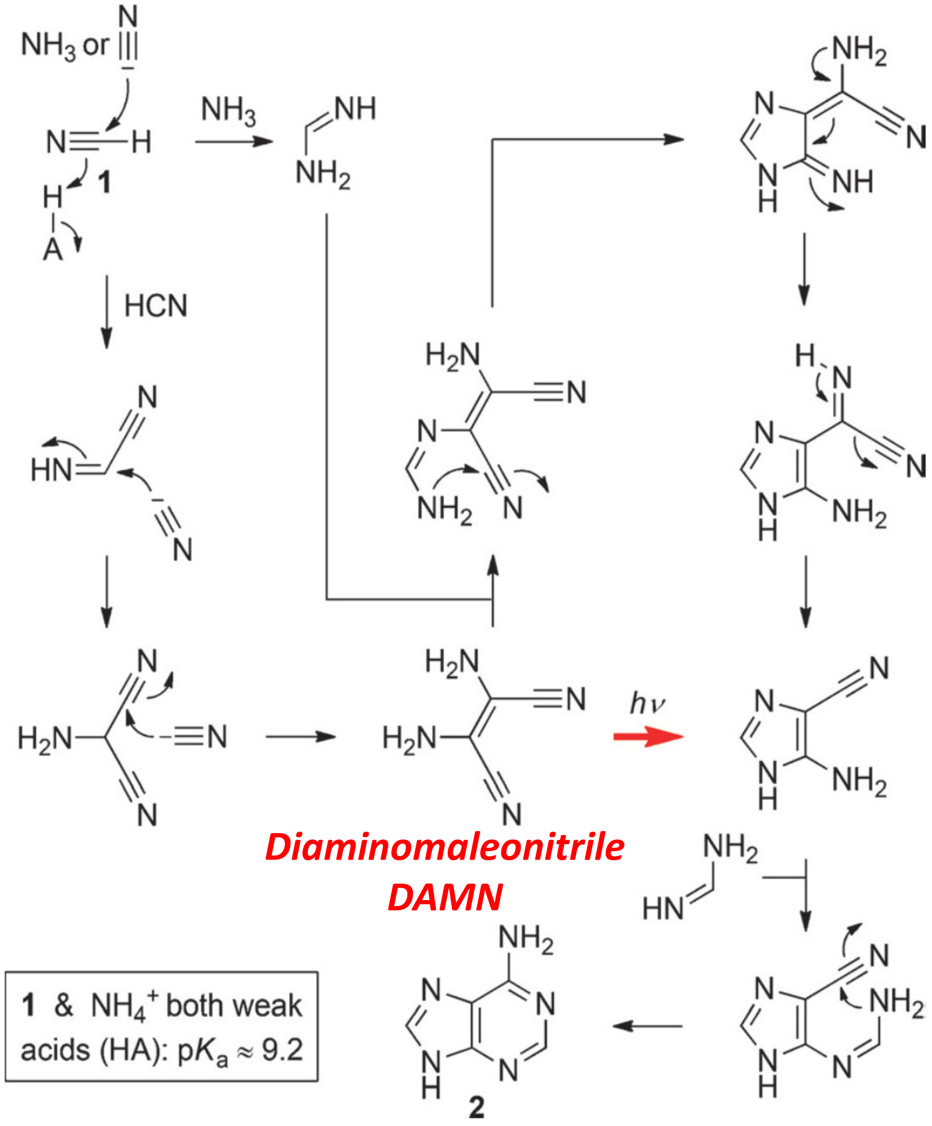
Powner, M. W.; Sutherland, J. D.; Szostak, J. W. *J. Am. Chem. Soc.* **2010**, *132*, 16677

T. Carell, *Nature* **2016**, *352(6287)*, 833-836

Prebiotic synthesis of nucleobases



Prebiotic synthesis of adenine – the pentamer of HCN!



Diaminomaleonitrile DAMN

1 & NH_4^+ both weak acids (HA): pK_a ≈ 9.2

1960 - Oró's synthesis of adenine **2** from hydrogen cyanide **1** and ammonia (general acid–base catalysis, presumed to operate in most steps, is only shown once).

Heating ammonium cyanide at 70^0C for a few days

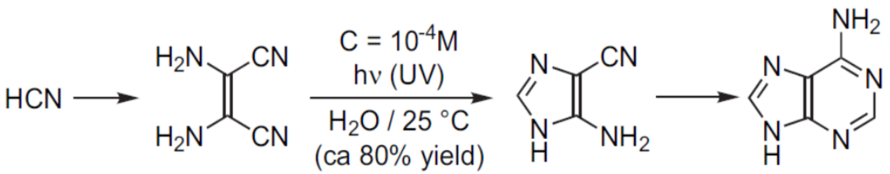
→ 0.5% adenine

Heating HCN with liquid ammonia in a sealed tube → 20% adenine

The photochemical shortcut discovered by Ferris and Orgel is shown by the red arrow.

Optimized yields – up to 20% for adenine, 3% for guanine

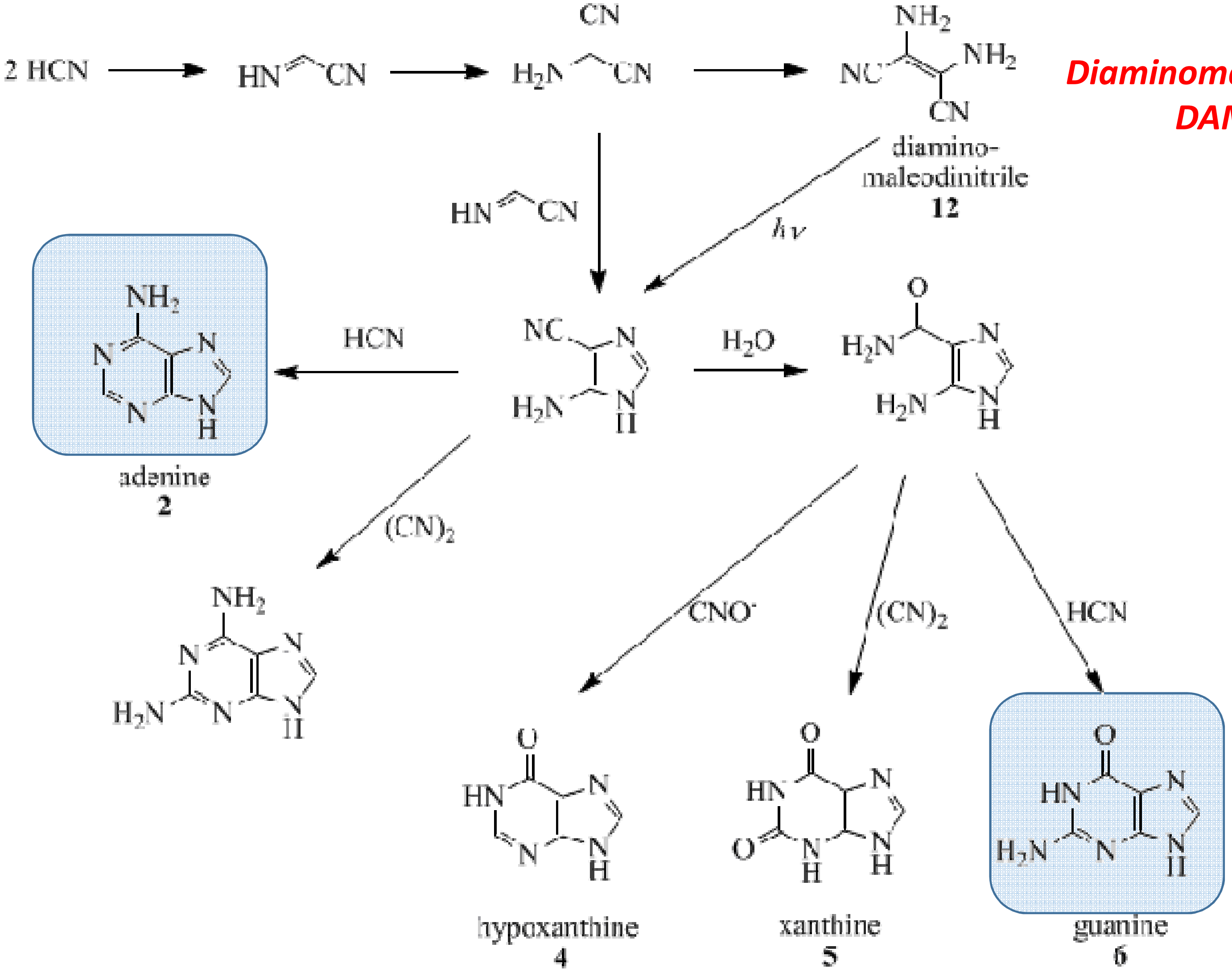
Eutectic freezing (-20^0C) increases the yield of DAMN formation by concentrating HCN between pure ice crystals



J. Oro *Biochem. Biophys. Res. Commun.* **1960**, *2*, 407.

J. P. Ferris, L. E. Orgel, *J. Am. Chem. Soc.* **1966**, *88*, 1074

Prebiotic synthesis of purines

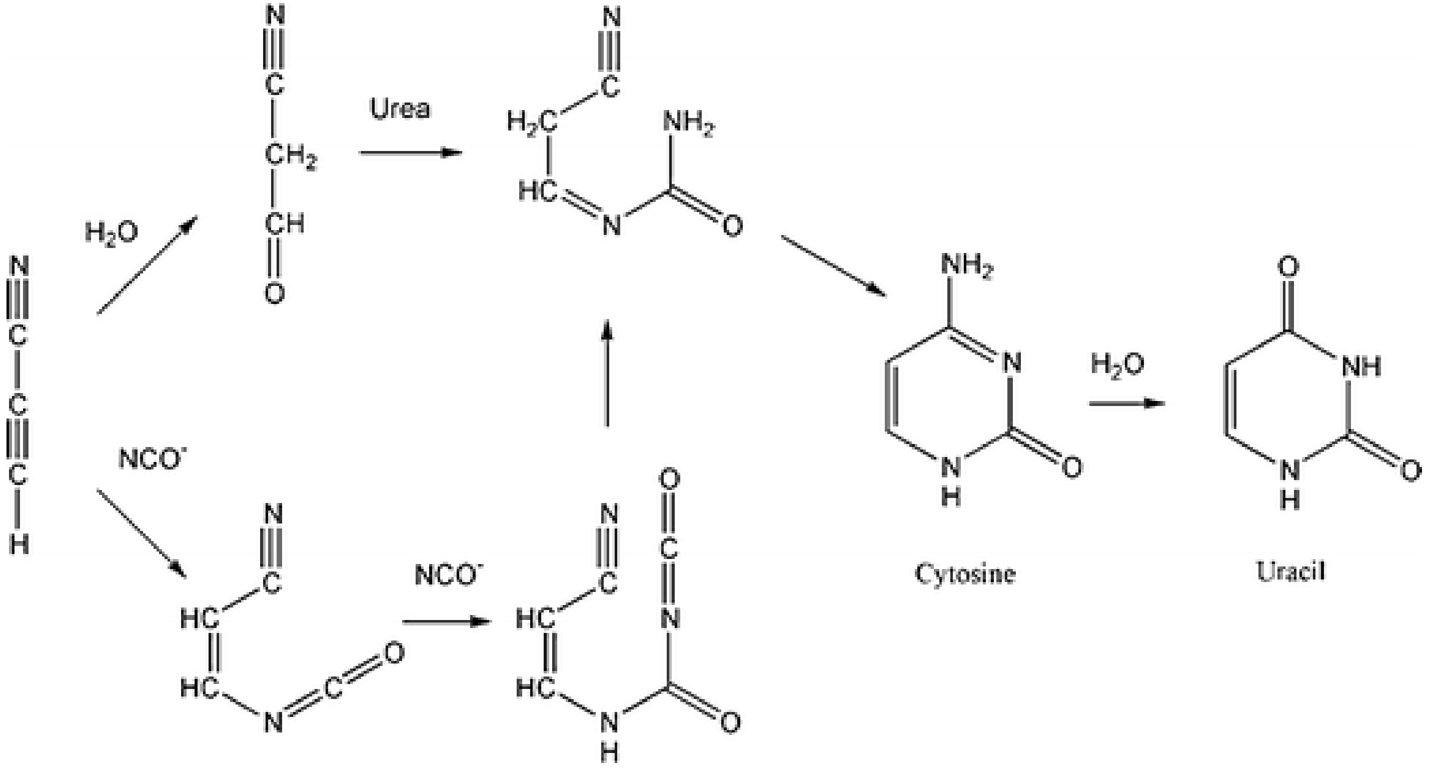


Diaminomaleonitrile
DAMN

Prebiotic synthesis of pyrimidines

Cyanoacetylene is a major product of electric discharges in the mixture of nitrogen and methane

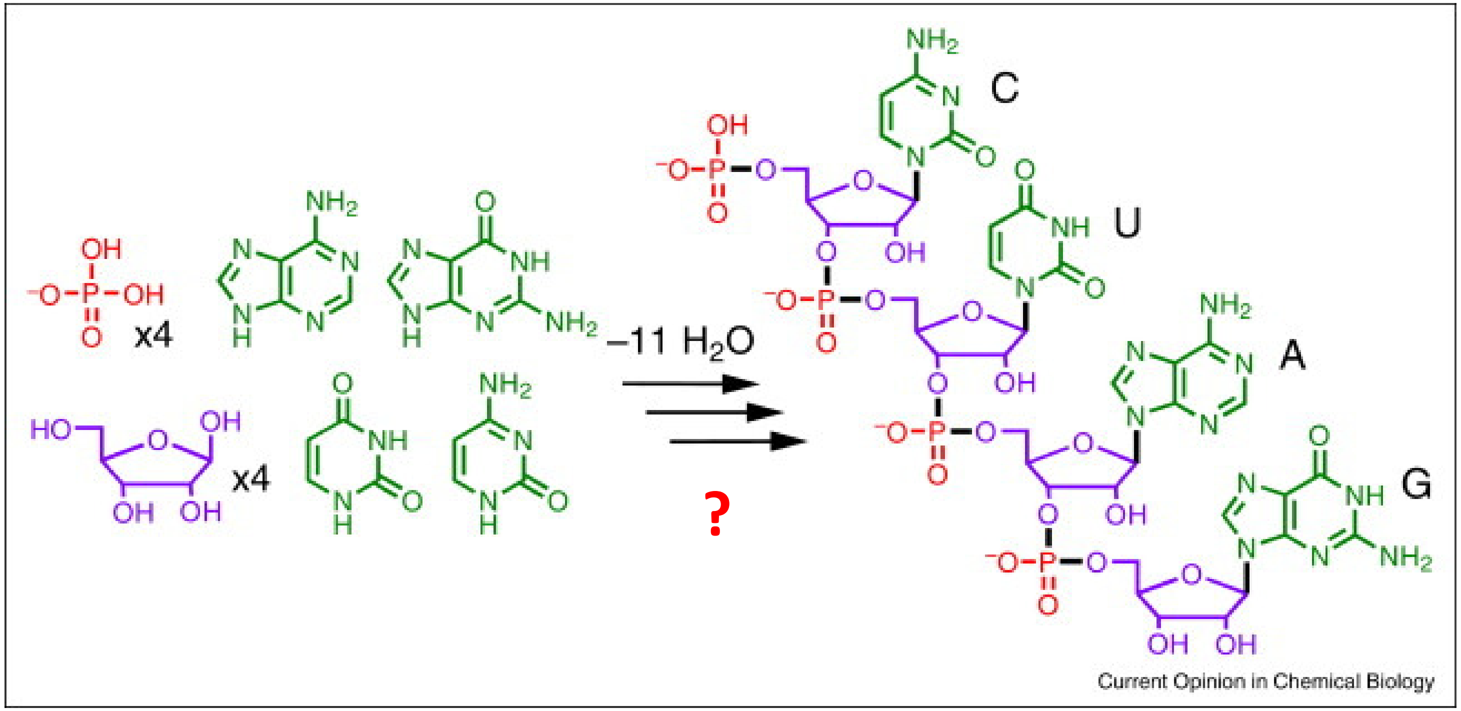
Cyanoacetylene can be hydrolysed to cyanoacetaldehyde. That compound can condense with urea to form cytosine



Cyanoacetylene incubated with saturated solution of urea yields up to 50% **cytosine**.
Other methods typically yield up to 5% cytosine.It is further converted to uracil by hydrolysis.

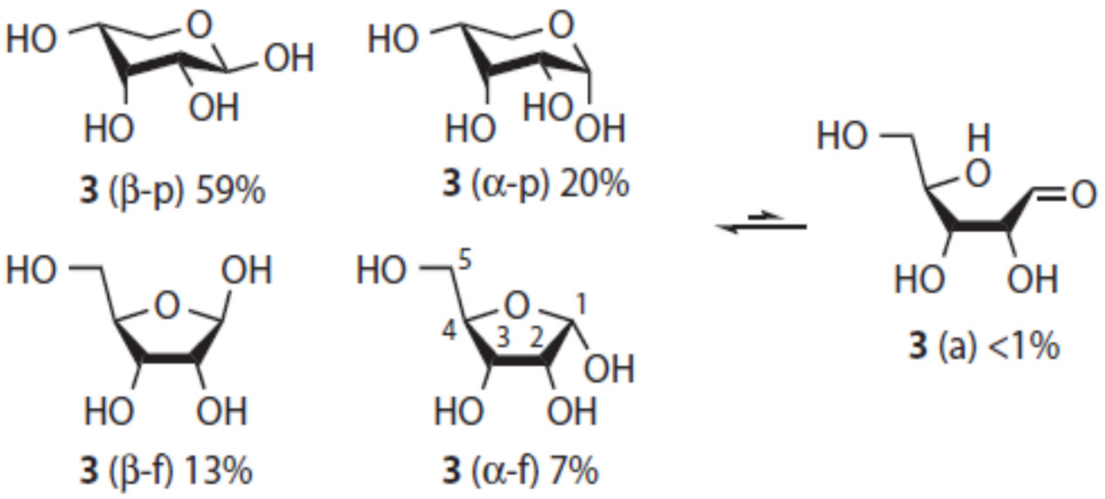
R. Shapiro PNAS **1999**, *96,* 4396-4401

Prebiotic synthesis of nucleosides, nucleotides and RNA



Current Opinion in Chemical Biology

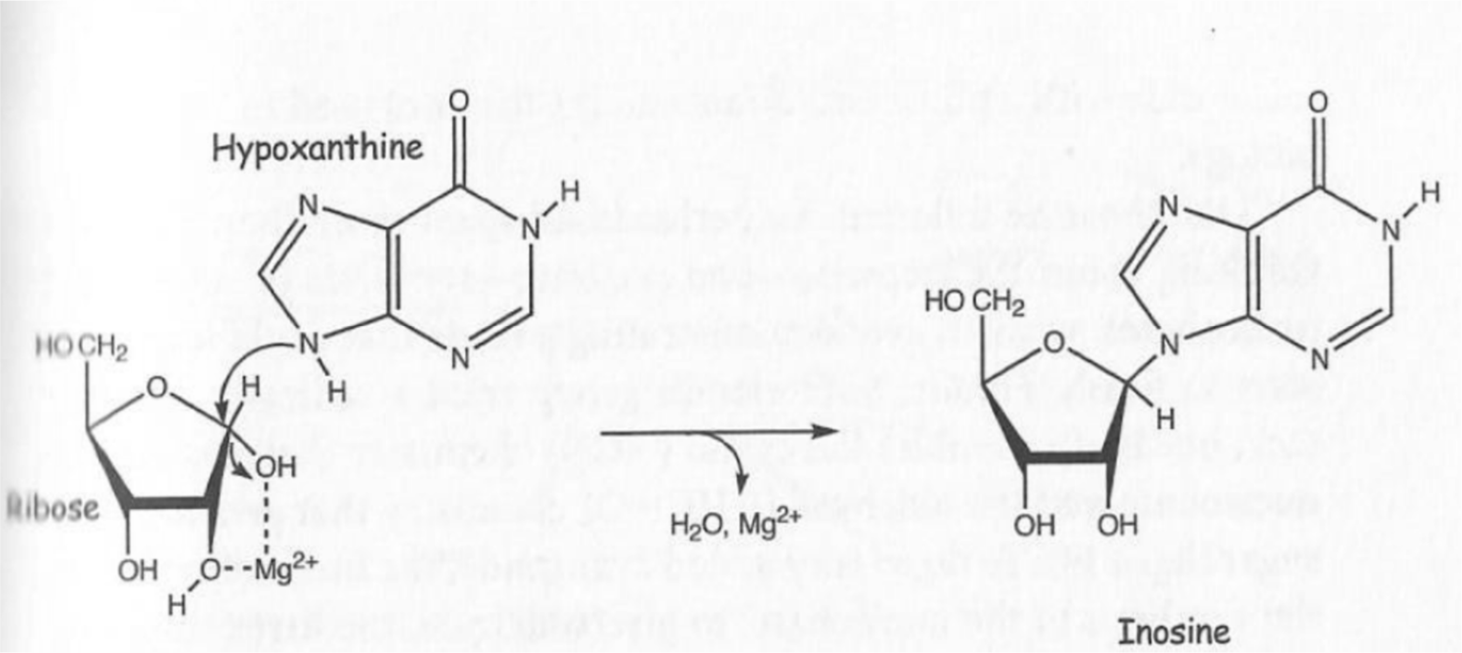
Prebiotic synthesis of nucleosides



The difficulties of assembling beta-ribonucleosides by nucleobase ribosylation:

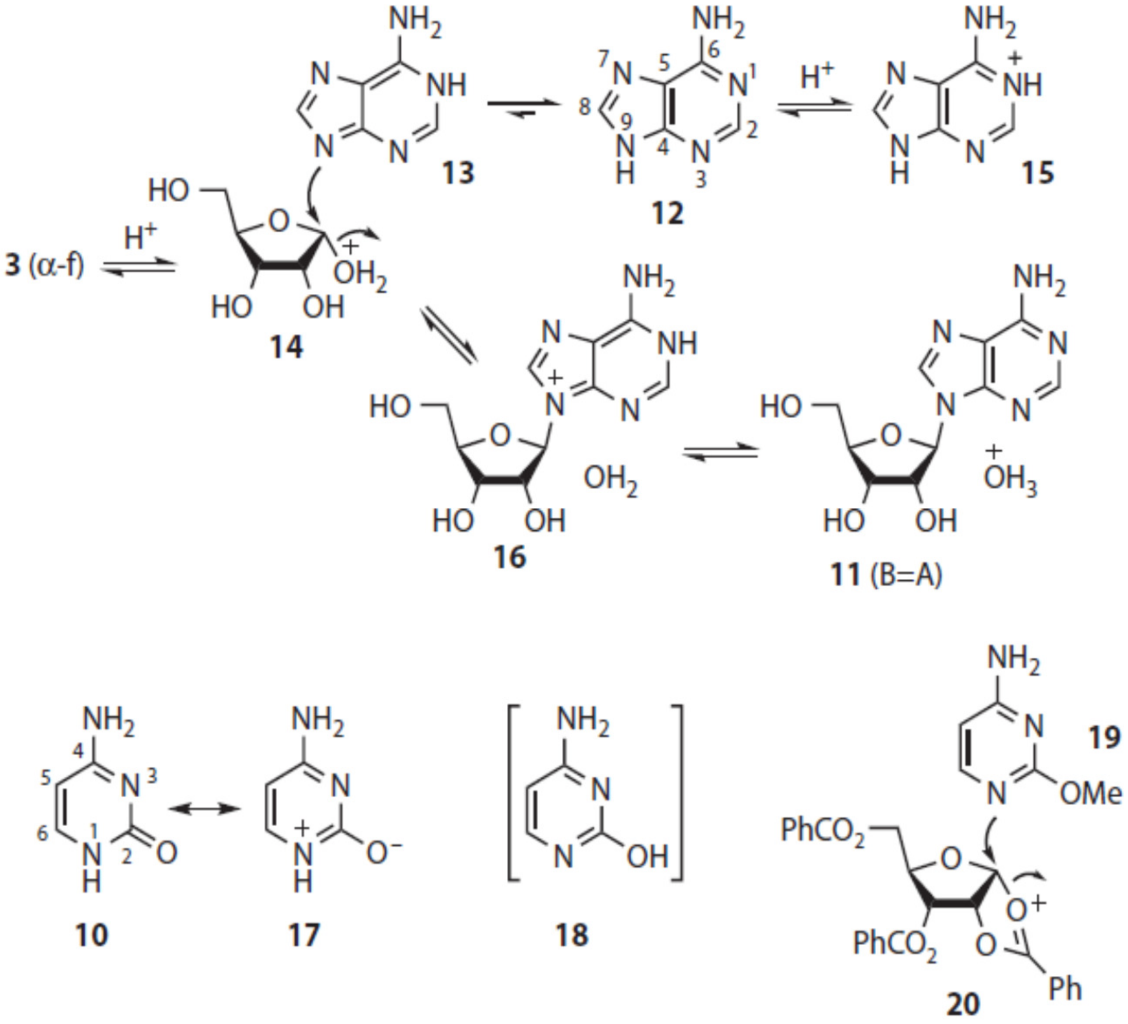
The many different forms of ribose **3** adopted in aqueous solution. The pyranose (p) and furanose (f) forms interconvert via the open-chain aldehyde (a), which is also in equilibrium with an open-chain aldehyde hydrate (not shown).

Prebiotic synthesis of nucleosides



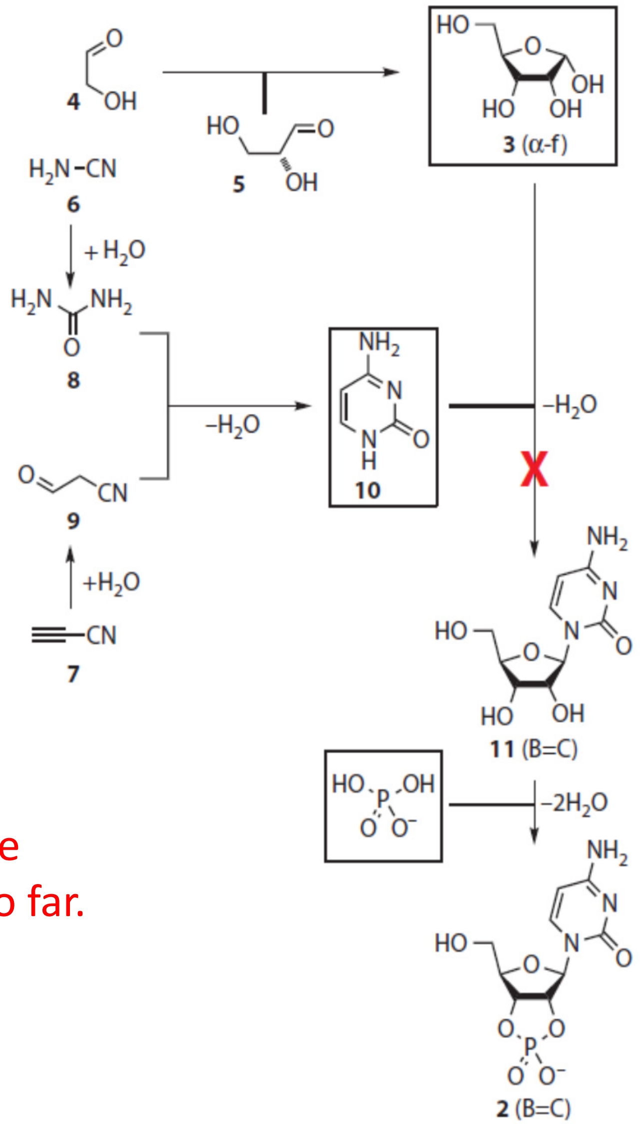
Heating purines with ribose and magnesium salts yields beta-nucleosides, although with low yields
β-inosine – 8%, β-adenosine – 4%, β-guanosine – 9%
Other isomers (e.g. alpha-glycosides) also present.

W. Fuller, R. Sanchez, L. Orgel *J. Mol. Biol.* **1972**, *67, 25-33*



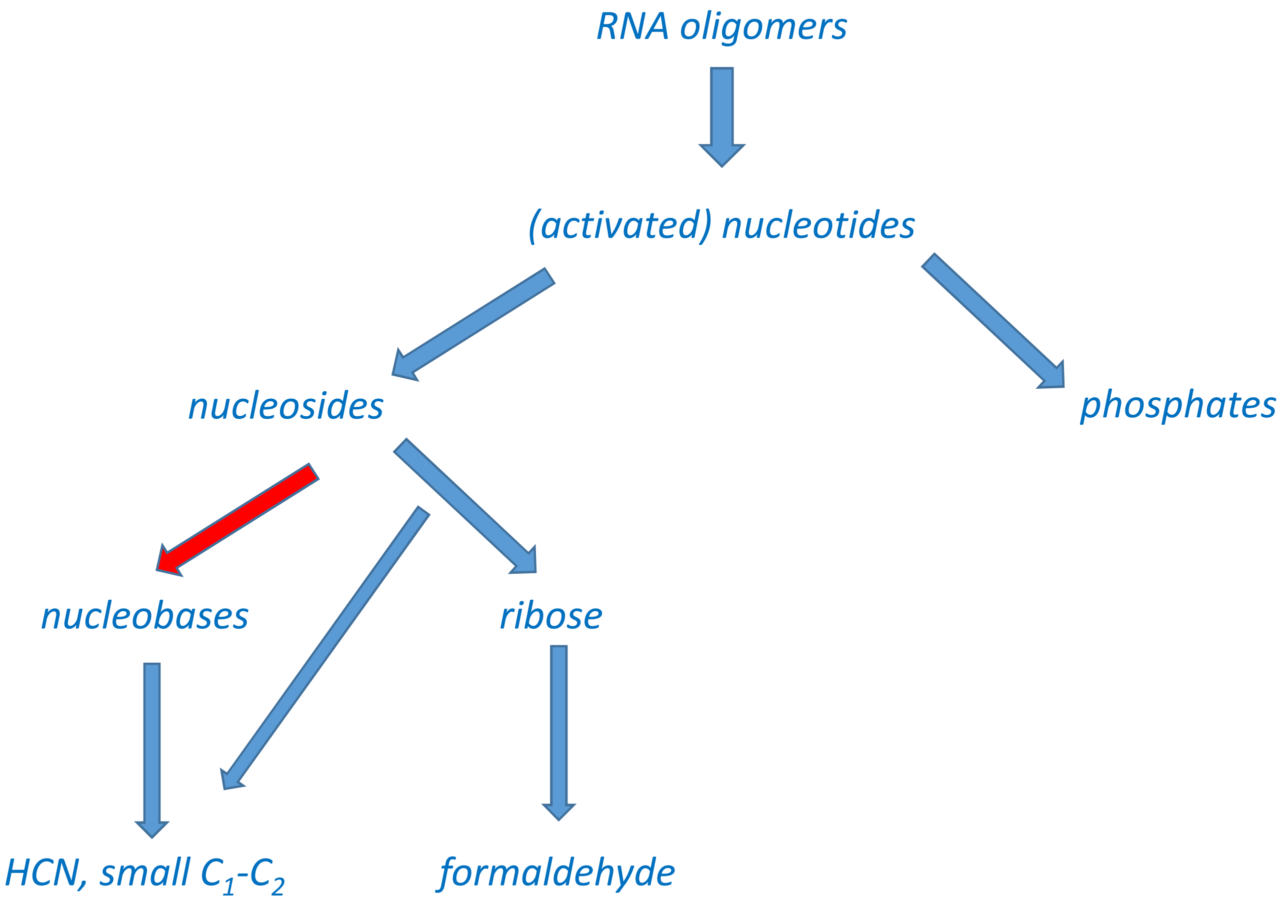
Adenine tautomerism and the ribosylation step necessary to make the adenosine **11** thought to be needed for RNA assembly. The low abundance of the reactive entities **13** and **14** is partly responsible for the low yield of **11**. The reason for the lower nucleophilicity of *N1* of the pyrimidines, and the conventional synthetic chemist's solution to the problems of ribosylation.

Prebiotic synthesis of pyrimidine nucleosides



No direct synthesis of pyrimidine nucleosides from ribose reported so far.

RNA oligomers – prebiotic disconnections



RNA oligomers

(activated) nucleotides

nucleosides

phosphates

nucleobases

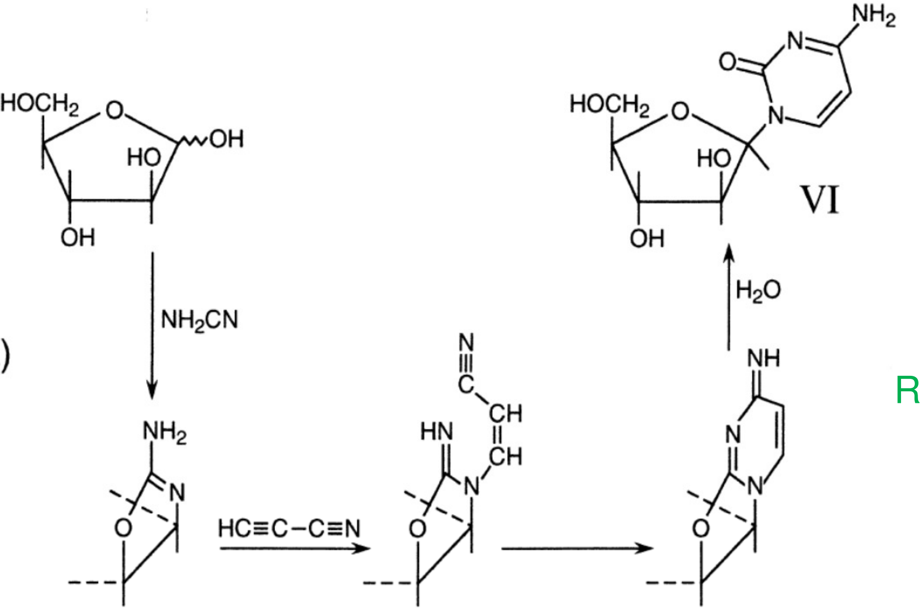
ribose

HCN, small C_1-C_2

formaldehyde

Prebiotic synthesis of nucleosides

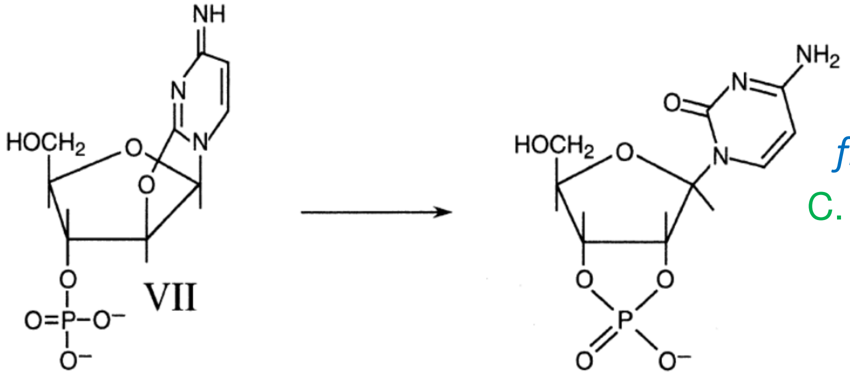
(a)



cytosine arabinoside synthesis

R. Sanchez, L. Orgel *J. Mol. Biol.* **1970**, *47, 531-543*

(b)



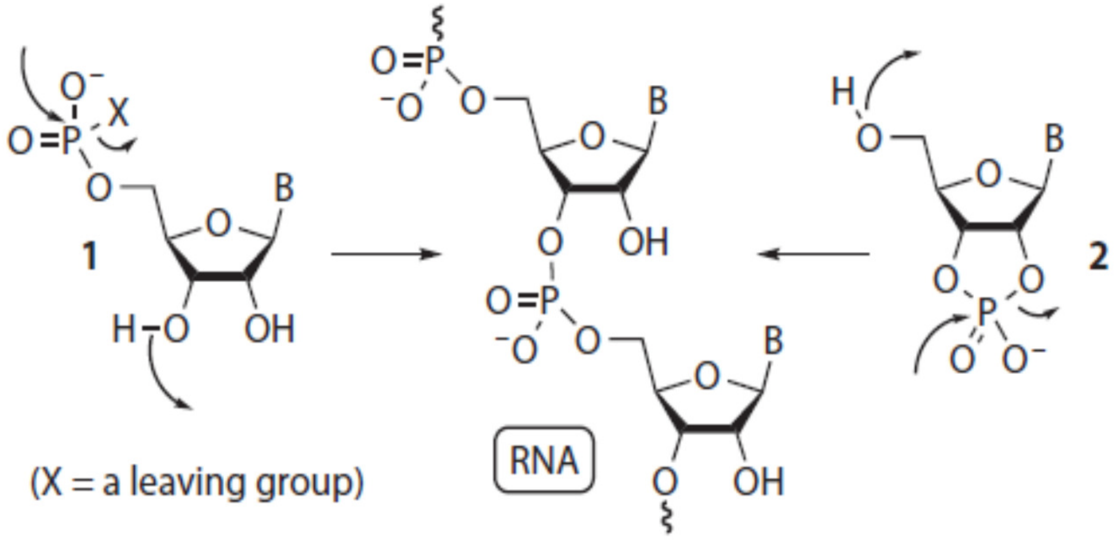
cytosine riboside (as a cyclic phosphate) is obtained from a phosphorylated substrate

C. M. Tapiero, J. Nagyvary *Nature* **1971**, *231, 42-43*

Review: L. E. Orgel *Crit. Rev. Biochem. Mol. Biol.* **2004**, *39, 99123*

Ara-3'P + NH_2CN + HCC-CN → Cyt-2',3'cP: A. Ingar, R. W. A. Luke, B. R. Hayter, J. D. Sutherland *ChemBioChem* **2003**, *4,* 504-507

Prebiotic synthesis of oligonucleotides

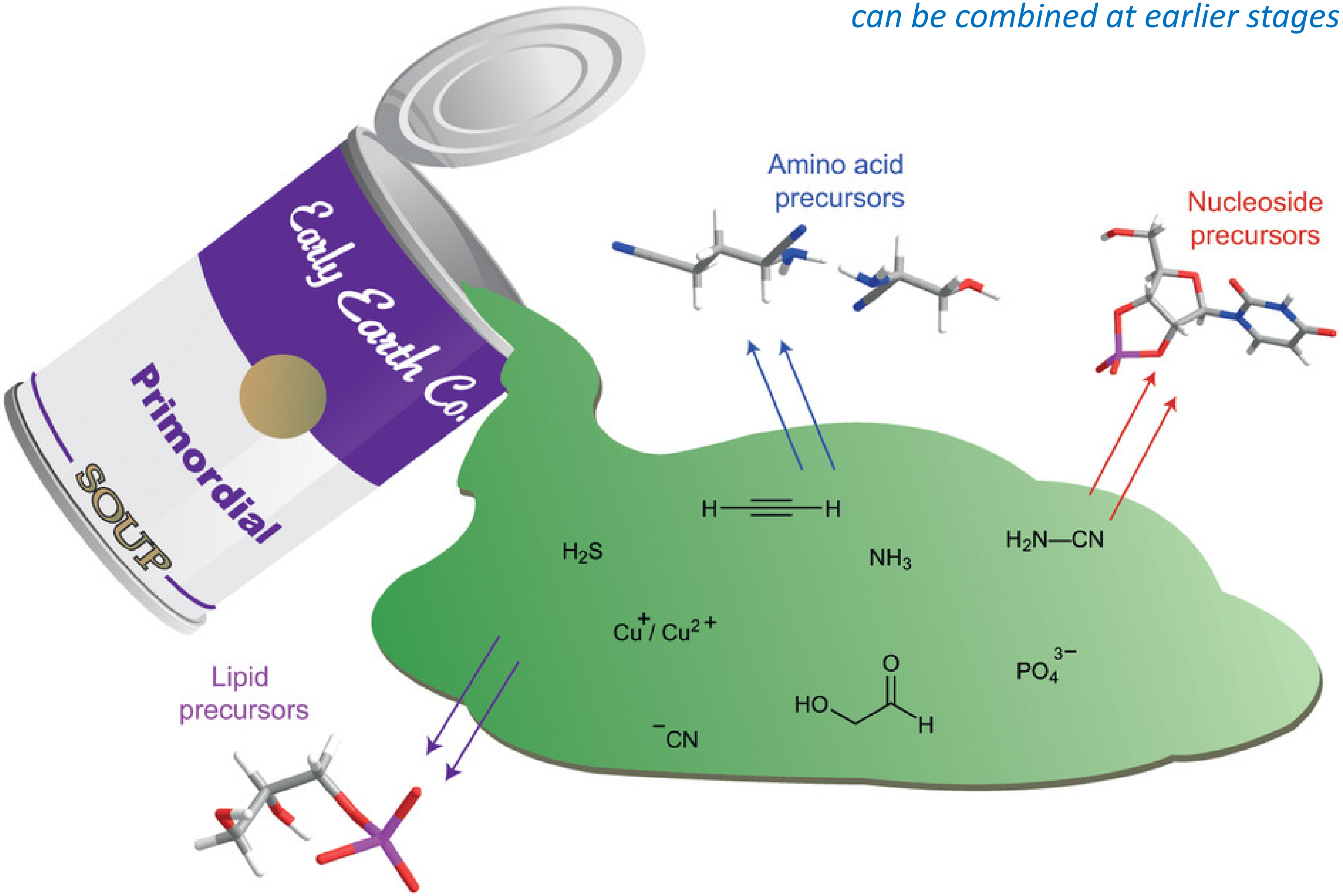


(X = a leaving group)

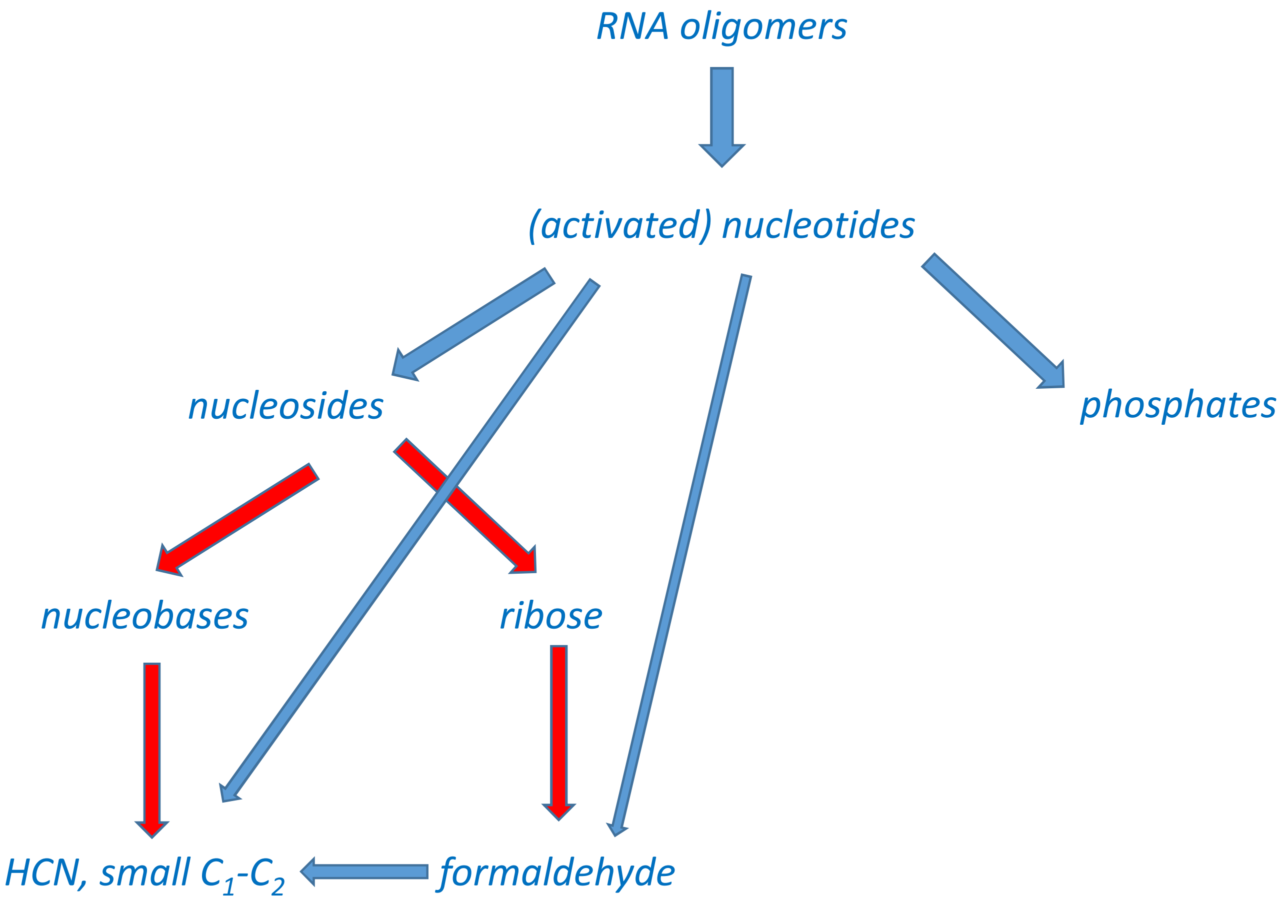
Activated ribonucleotides in the potentially prebiotic assembly of RNA. Potential P–O bond forming polymerization chemistry is indicated by the curved arrows.

Cyanosulfidic chemistry

The aldol chemistry of sugars and cyanide chemistry of nucleobases can be combined at earlier stages than glycosylation.



RNA oligomers – prebiotic disconnections



RNA oligomers

(activated) nucleotides

nucleosides

phosphates

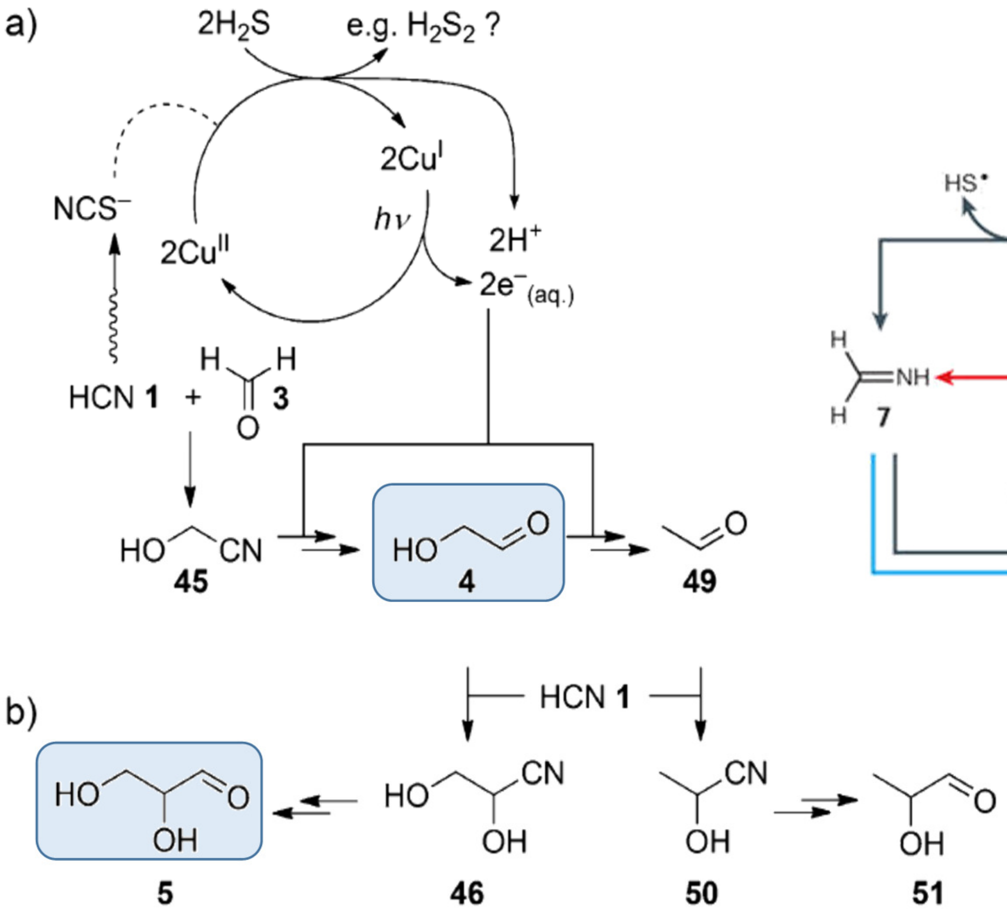
nucleobases

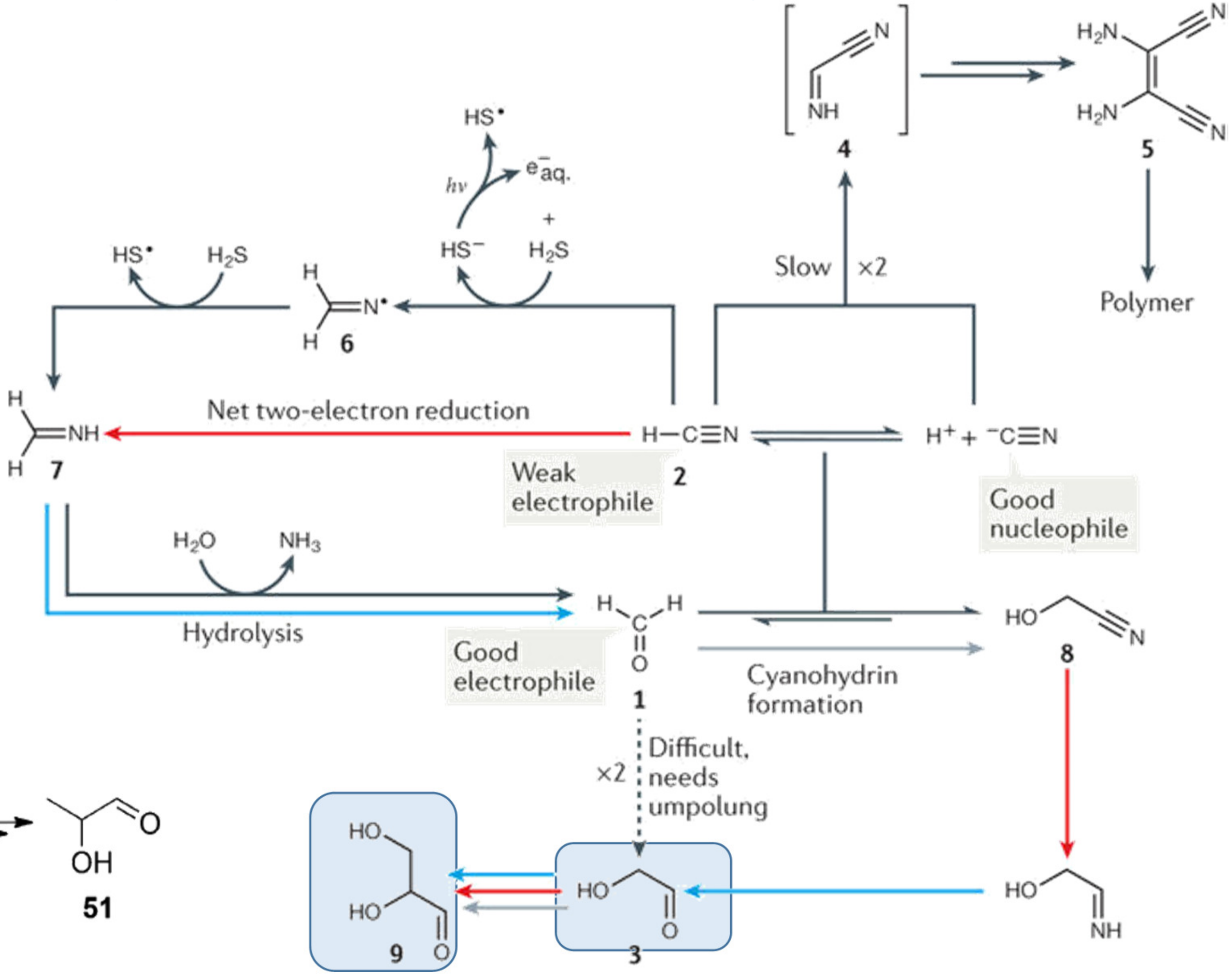
ribose

HCN, small C_1-C_2

formaldehyde

Cyanosulfidic chemistry for the Kiliani-Fischer homologation

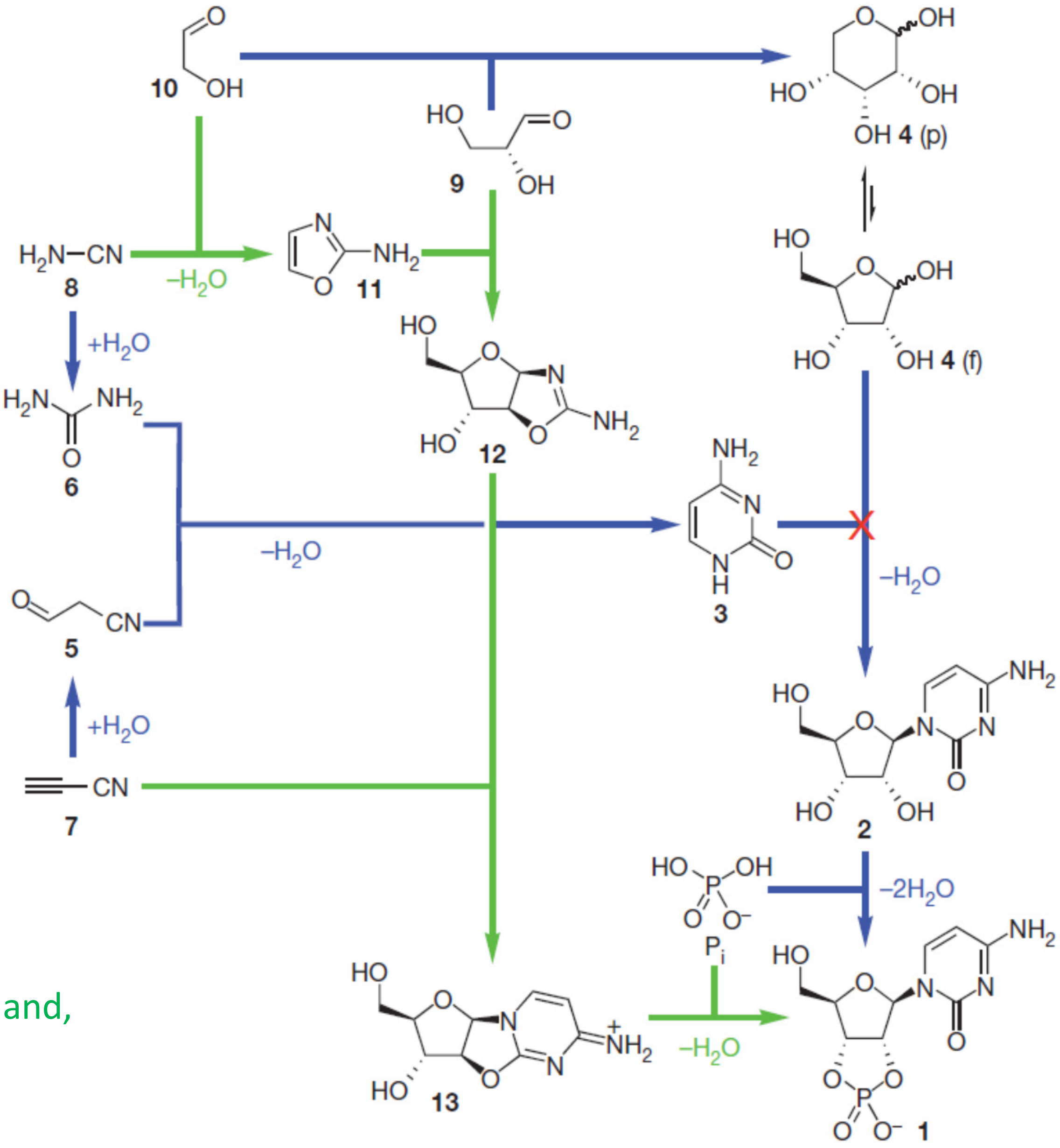




Nature Reviews | Chemistry

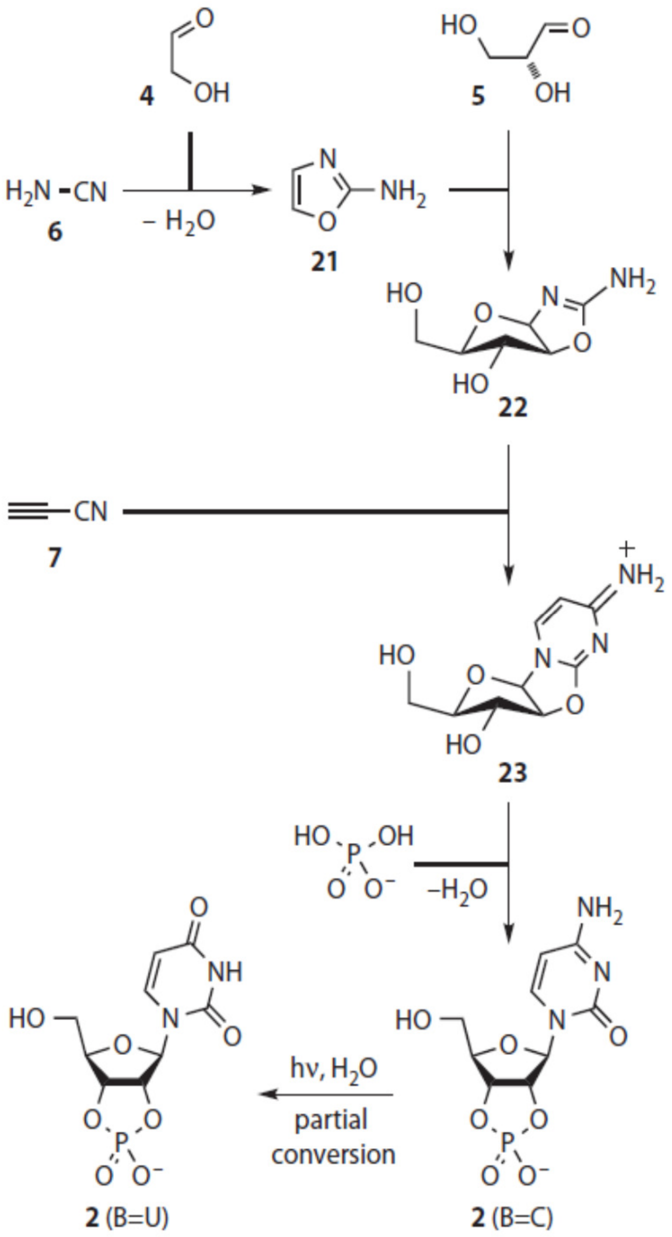
J. Sutherland, *Nature Reviews Chemistry* **2017**, *1*, Article 0012, doi:10.1038/s41570-016-0012

Cyanosulfidic chemistry



M. W. Powner, B. Gerland, J. D. Sutherland,
Nature **2009**, *459*, 239–242

Cyanosulfidic chemistry

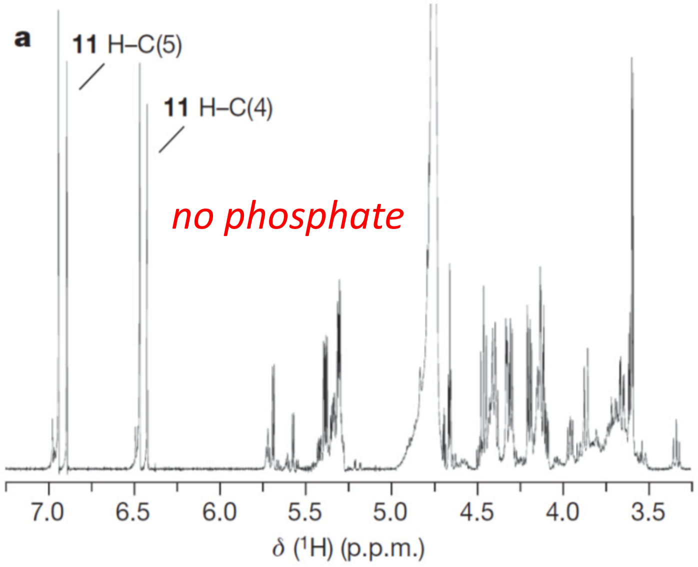


The recently uncovered route to activated pyrimidine nucleotides **2**.

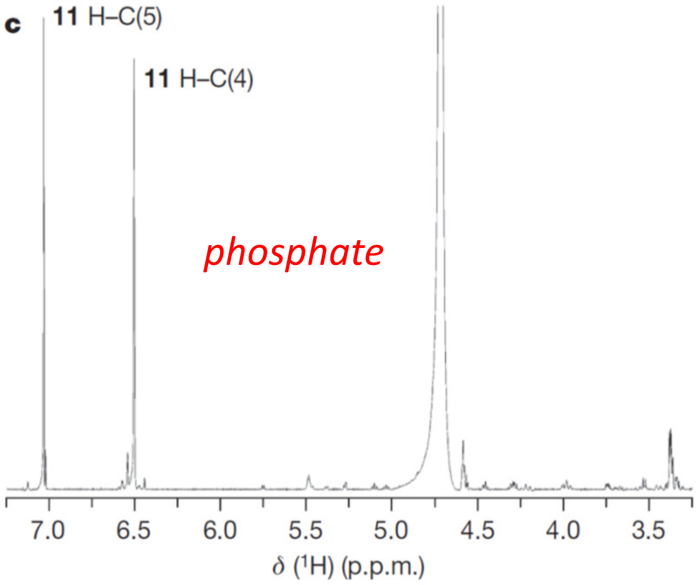
The nucleobase ribosylation problem is circumvented by the assembly proceeding through 2-aminooxazole **21**, which can be thought of as the chimera of half a pentose sugar and half a nucleobase. The second half of the pentose - glyceraldehyde **5** -and the second half of the nucleobase—cyanoacetylene **7**—are then added sequentially to give the anhydronucleoside **23**.

Phosphorylation and rearrangement of **23** then furnishes **2** (B=C), and UV irradiation effects the partial conversion of **2** (B=C) to **2** (B=U).

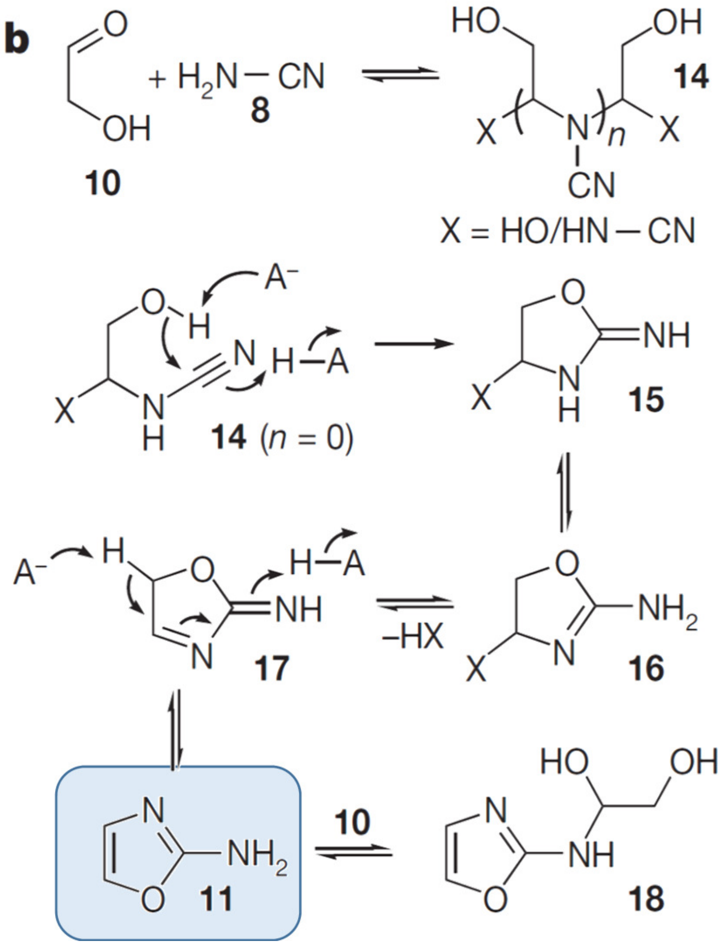
Cytosine-2’,3’cP – step 1: 2-amino-oxazole

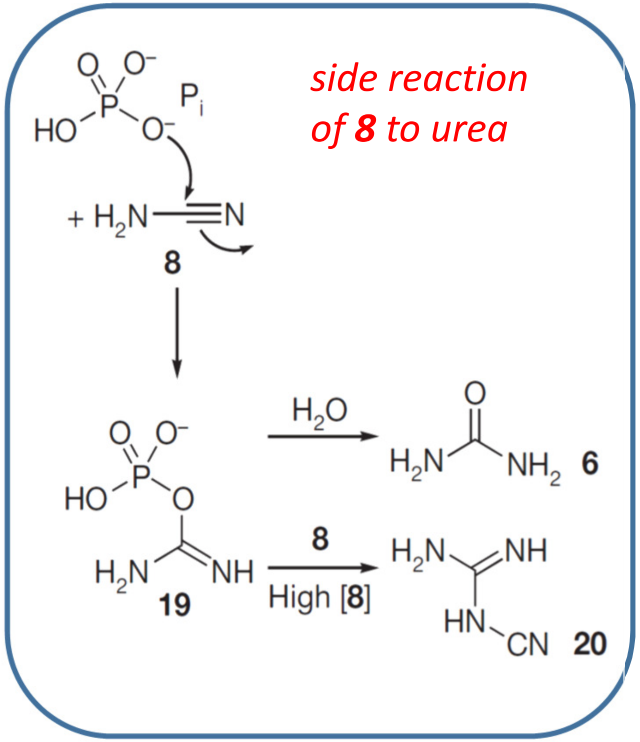


no phosphate



phosphate

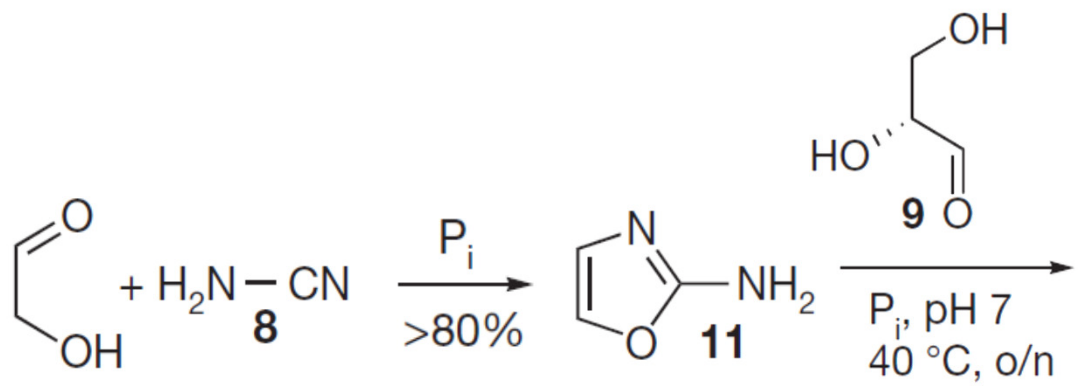




*side reaction of **8** to urea*

M. W. Powner, B. Gerland, J. D. Sutherland,
Nature **2009**, *459*, 239–242

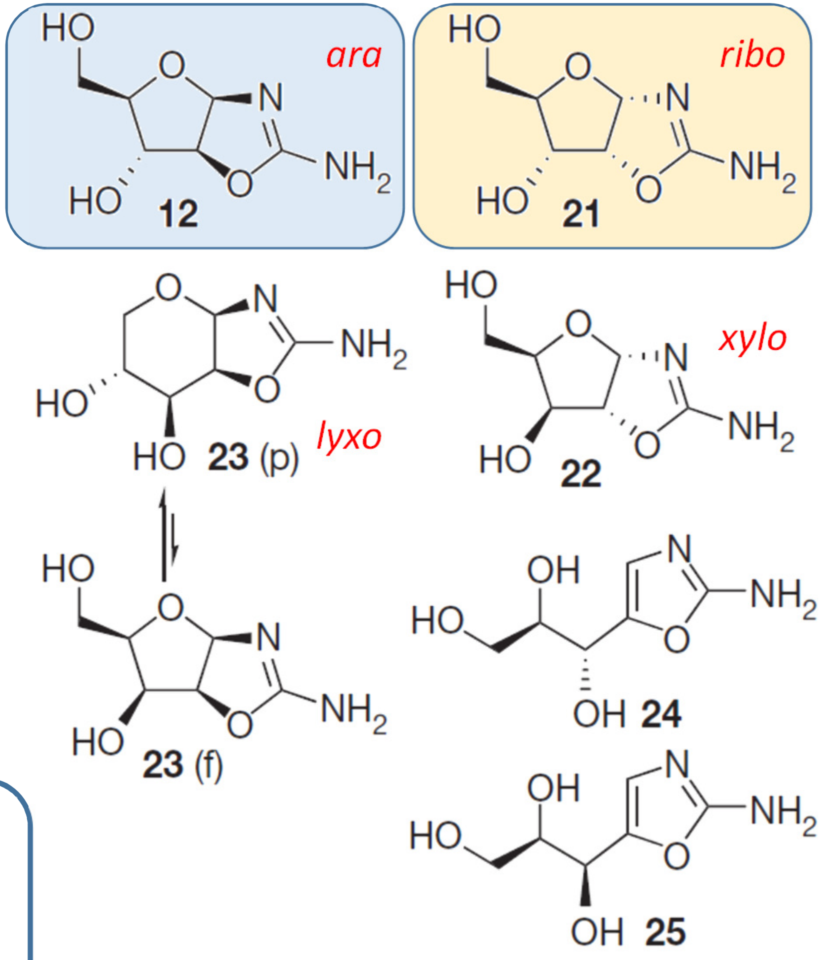
Cytosine-2',3'cP – step 2: pentose-amino-oxazolines

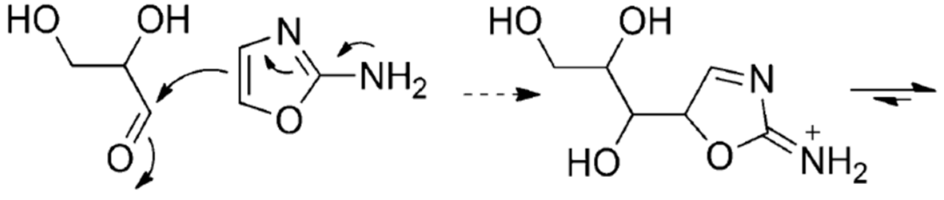


12 15%
21 25%
22 6%
23 4%
24 11%
25 9%

Yields for two steps based on **10**

*ribo-**21** – selectively crystallizes on cooling → ara-**12** becomes the main mixture component*



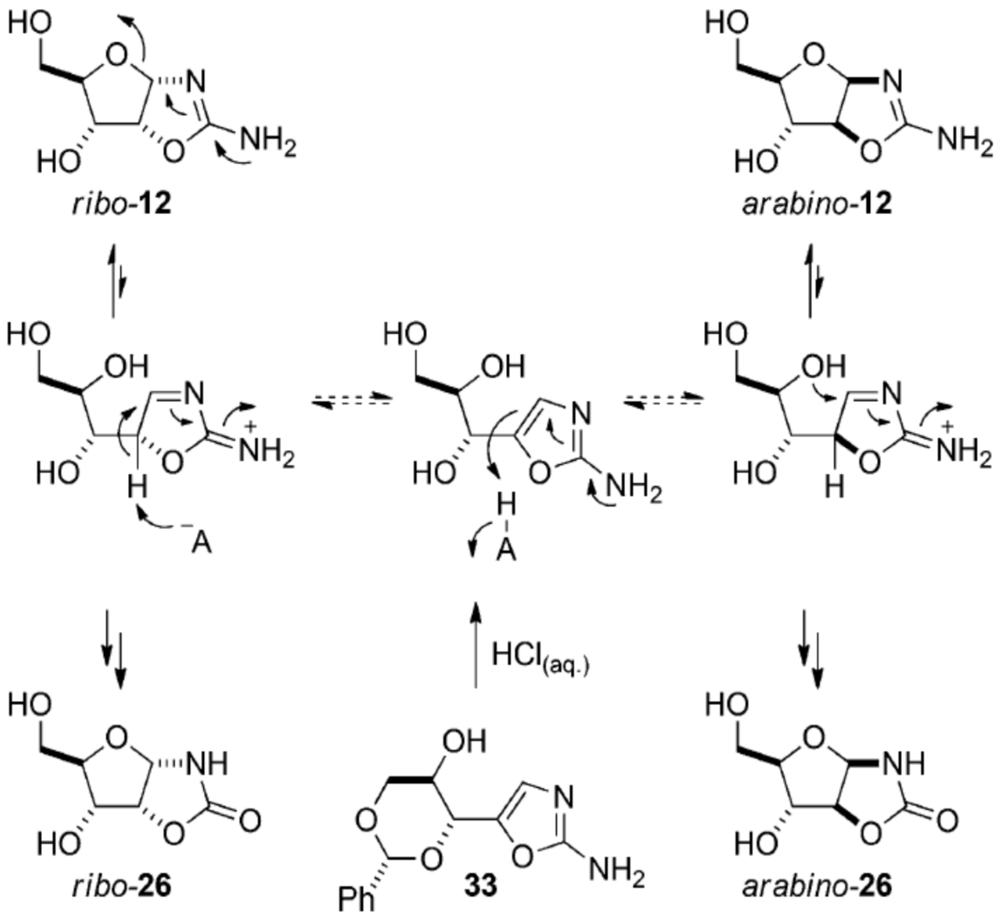


44% *ribo*-**12**
30% *arabino*-' **21**
13% *lyxo*- **23**
8% *xylo*- **22**

M. W. Powner, B. Gerland, J. D. Sutherland, *Nature* **2009**, *459*, 239–242

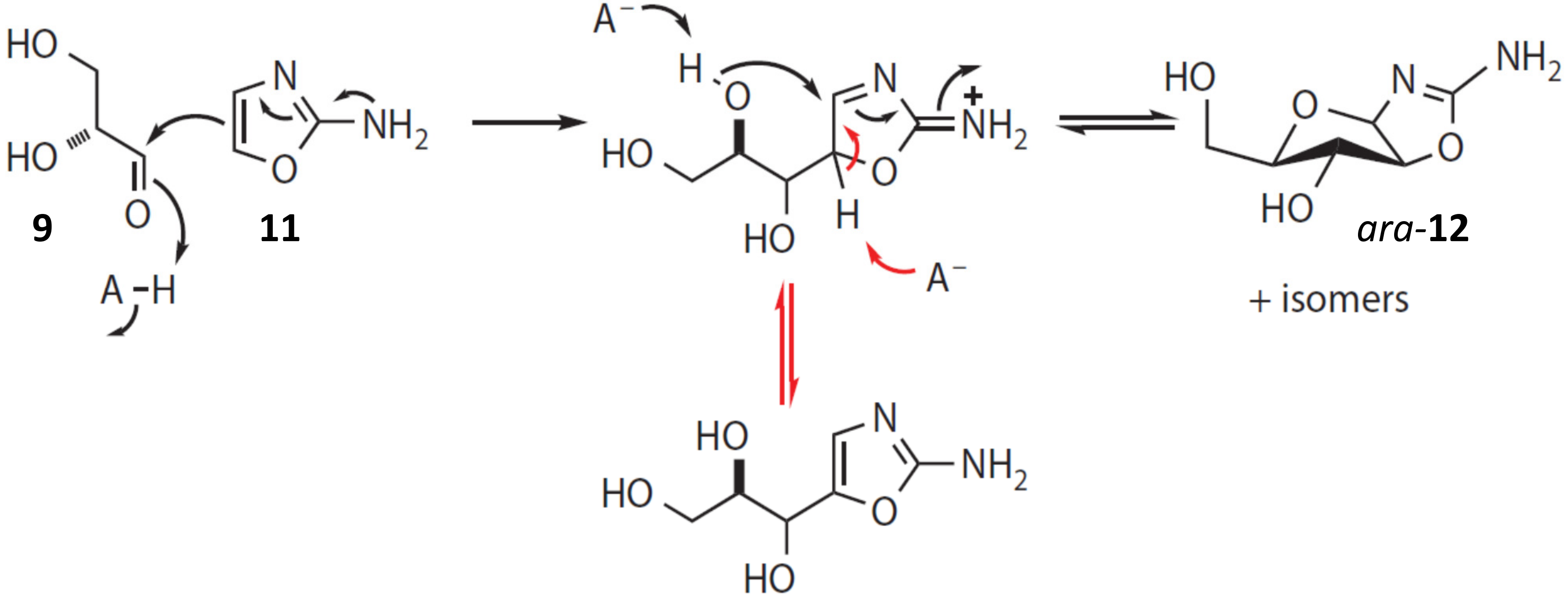
Cyanosulfidic chemistry

Interconversion of pentose aminooxazoline stereoisomers.

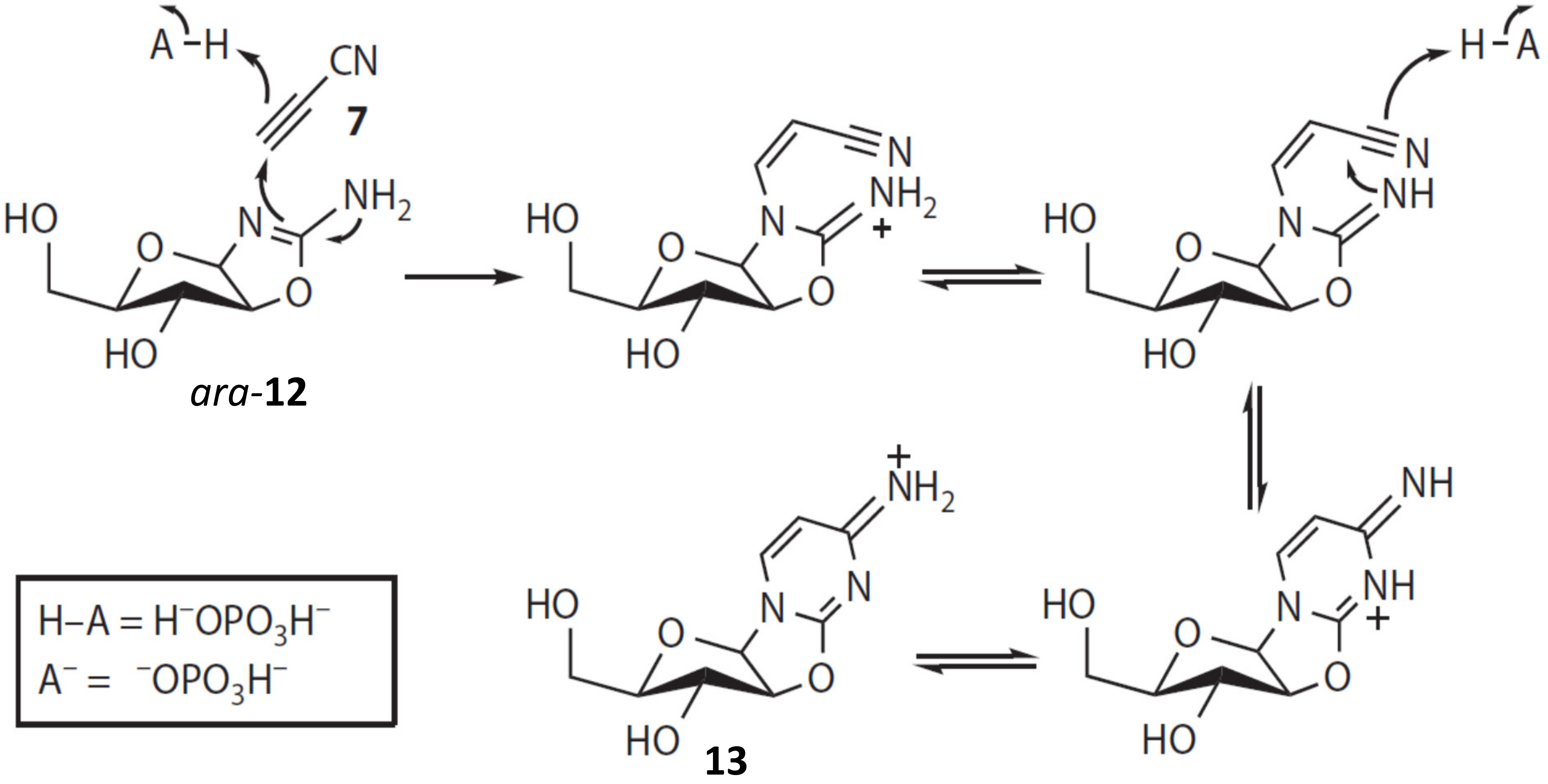


The „side product" *ribo*-**21** can be converted to the expected *arabino*-**12** by general acid-base catalysis in phosphate buffer, although with some decomposition to **26**

Cytosine-2’,3’cP – step 2: pentose-amino-oxazolines

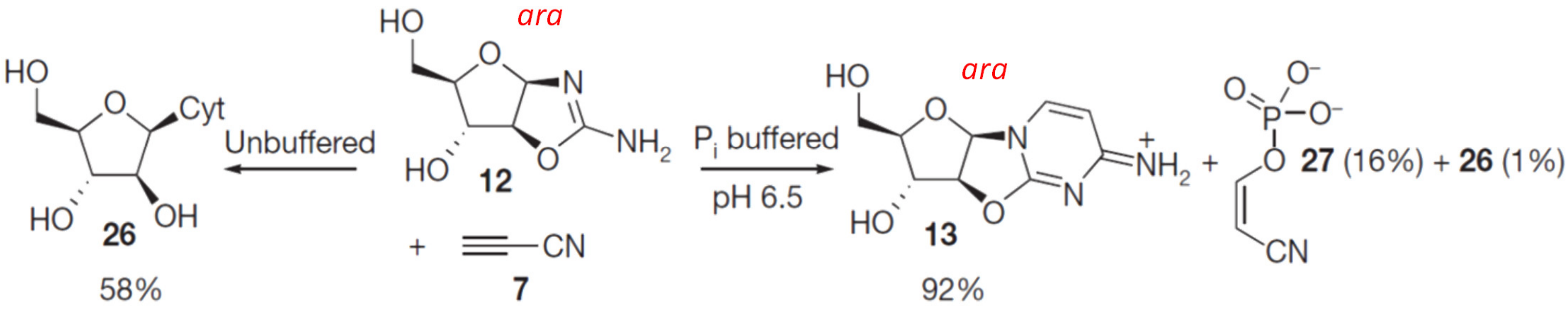


Cytosine-2’,3’cP – step 3: arabinose-anhydronucleosides



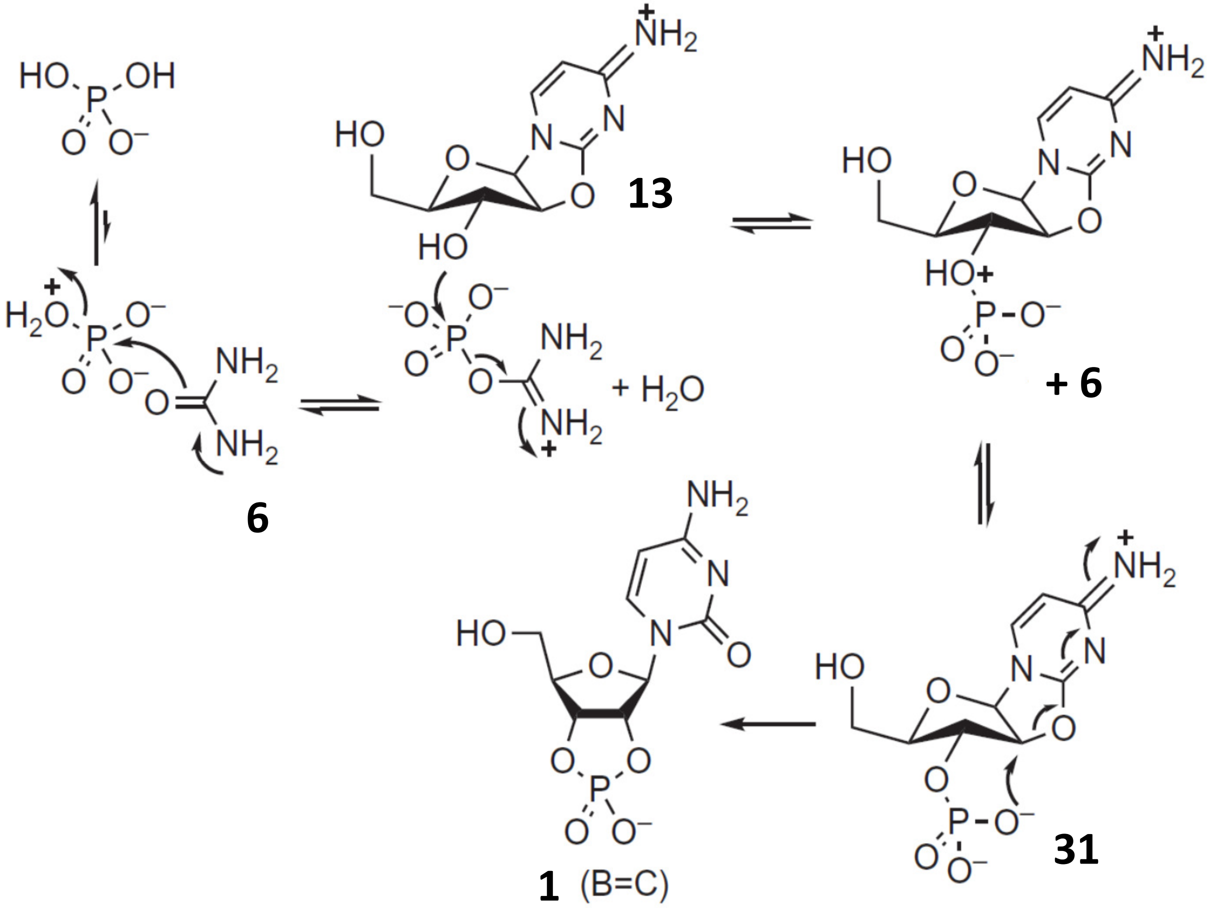
H–A = $H^{-}OPO_3H^{-}$
A^{-} = $^{-}OPO_3H^{-}$

Cytosine-2',3'cP – step 3: arabinose-anhydronucleosides



M. W. Powner, B. Gerland, J. D. Sutherland, *Nature* **2009**, *459*, 239–242

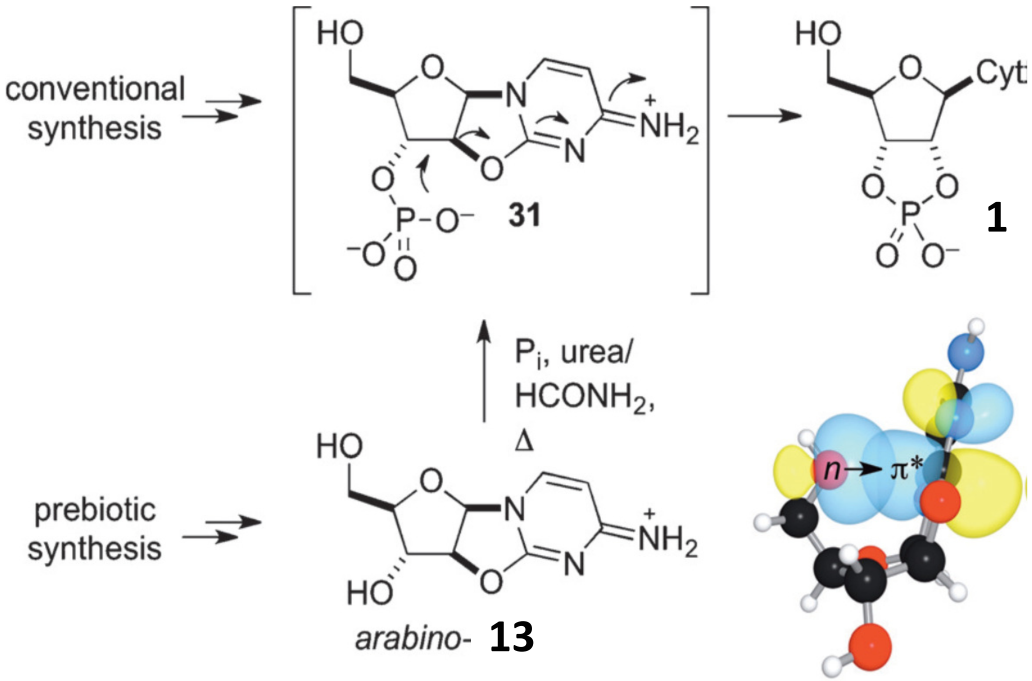
Cytosine-2',3'cP – step 4: *β*-ribocytidine-2',3'-cyclic phosphate



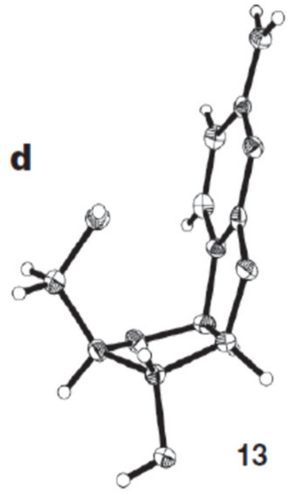
M. W. Powner, B. Gerland, J. D. Sutherland, *Nature* **2009**, *459*, 239–242

Cytosine-2’,3’cP – step 4: β-ribocytidine-2’,3’-cyclic phosphate

Rearrangement of **31**, the 3’-phosphate of *arabino*-**13**, to **1** by intramolecular nucleophilic substitution.

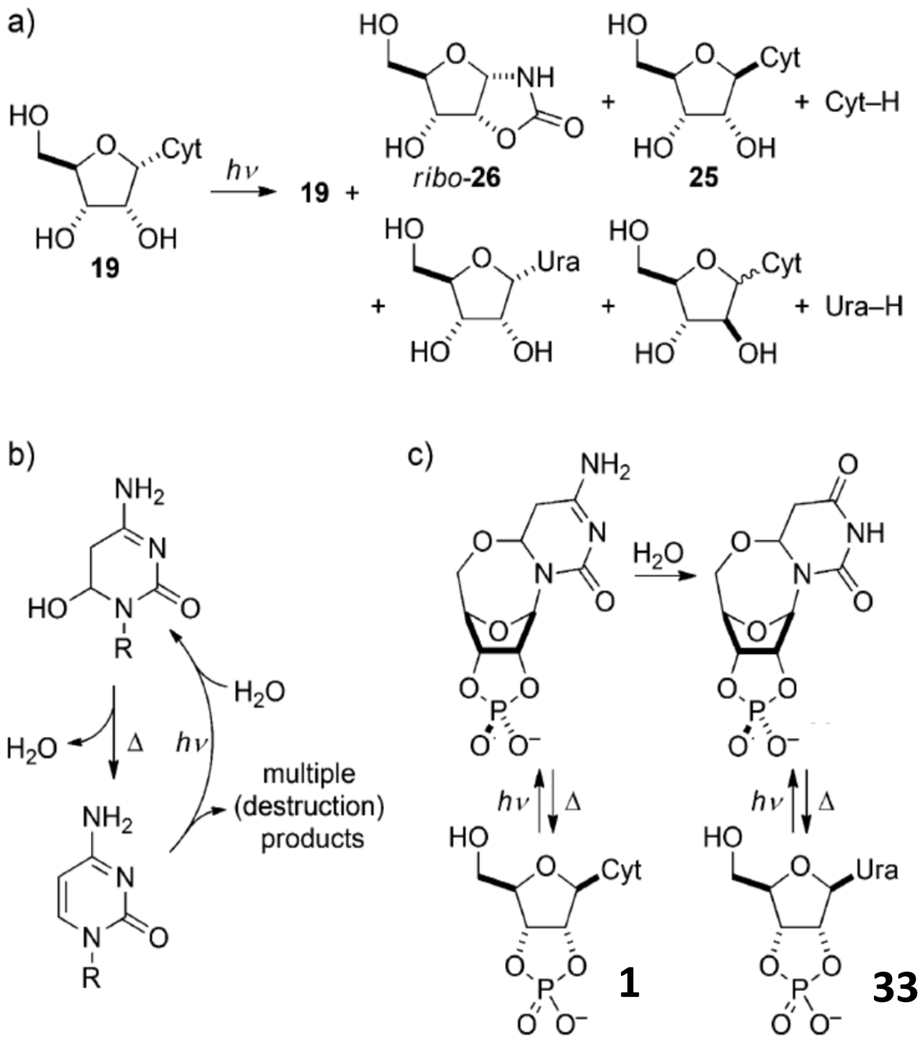


arabino- **13**



M. W. Powner, B. Gerland, J. D. Sutherland, *Nature* **2009**, *459*, 239–242

Activated pyrimidine nucleotides – step 5: UV-light induced rearrangement C→U + cleanup of the side products



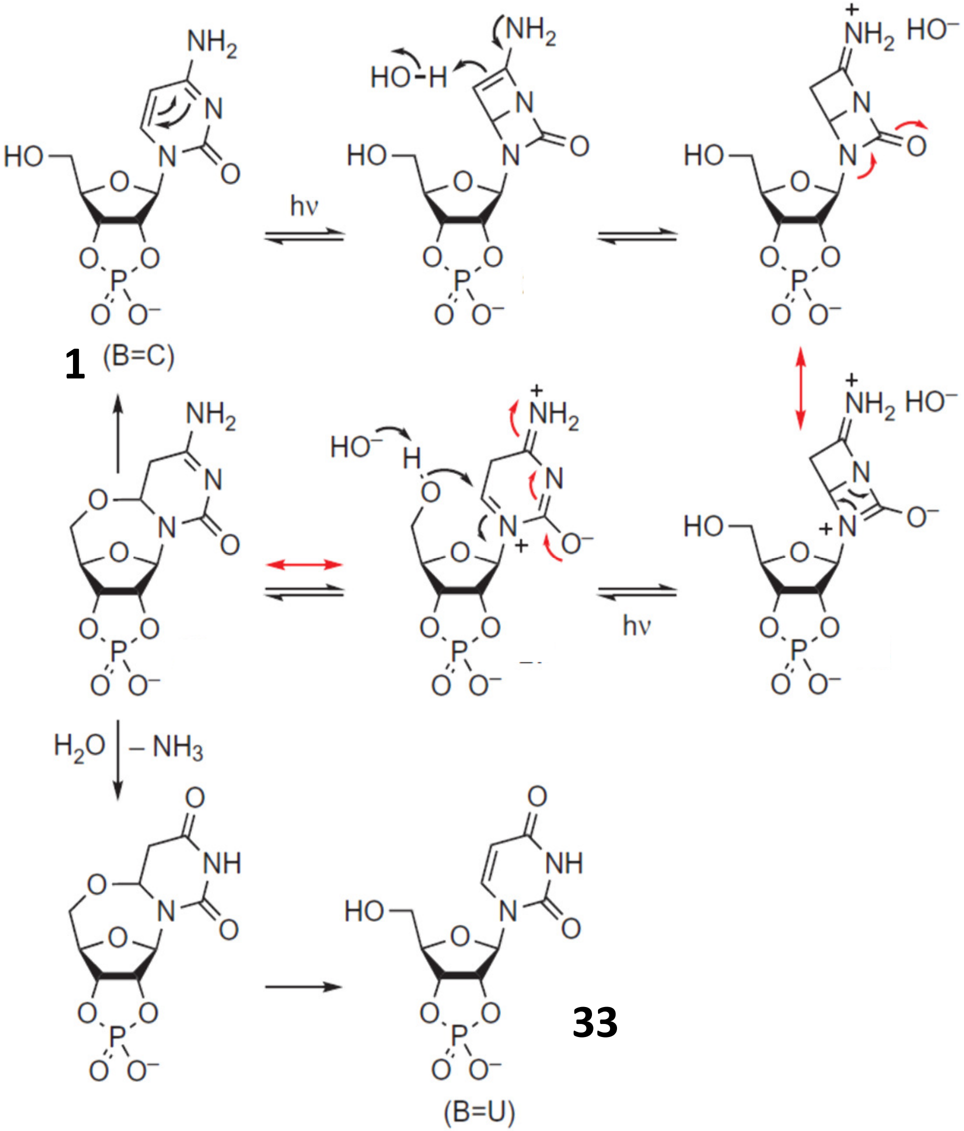
Orgel reported photoanomerization of alpha-cytidine **19** to the beta-anomer **25**, but the reaction was very low-yielding and combined with massive decomposition

R. A. Sanchez, L. E. Orgel, *J. Mol. Biol.* **1970**, *47,* 531-543

UV light tends to destroy most of phosphorylated cytidine derivatives (by photohydration and followed decomposition).
EXCEPTION: beta-cytidine-2’,3’-cyclic phosphate **1**
→ Partial conversion to the uridine nucleotide **33**, but both are stable on further irradiation

Photochemistry of cytidine nucleosides and nucleotides.

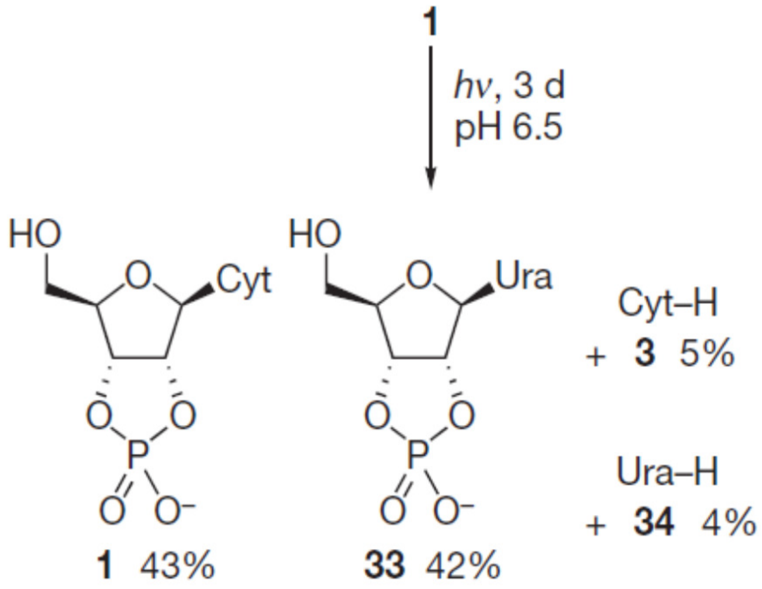
step 5: UV-light induced rearrangement C→U + cleanup of the side products



1 (B=C)

33 (B=U)

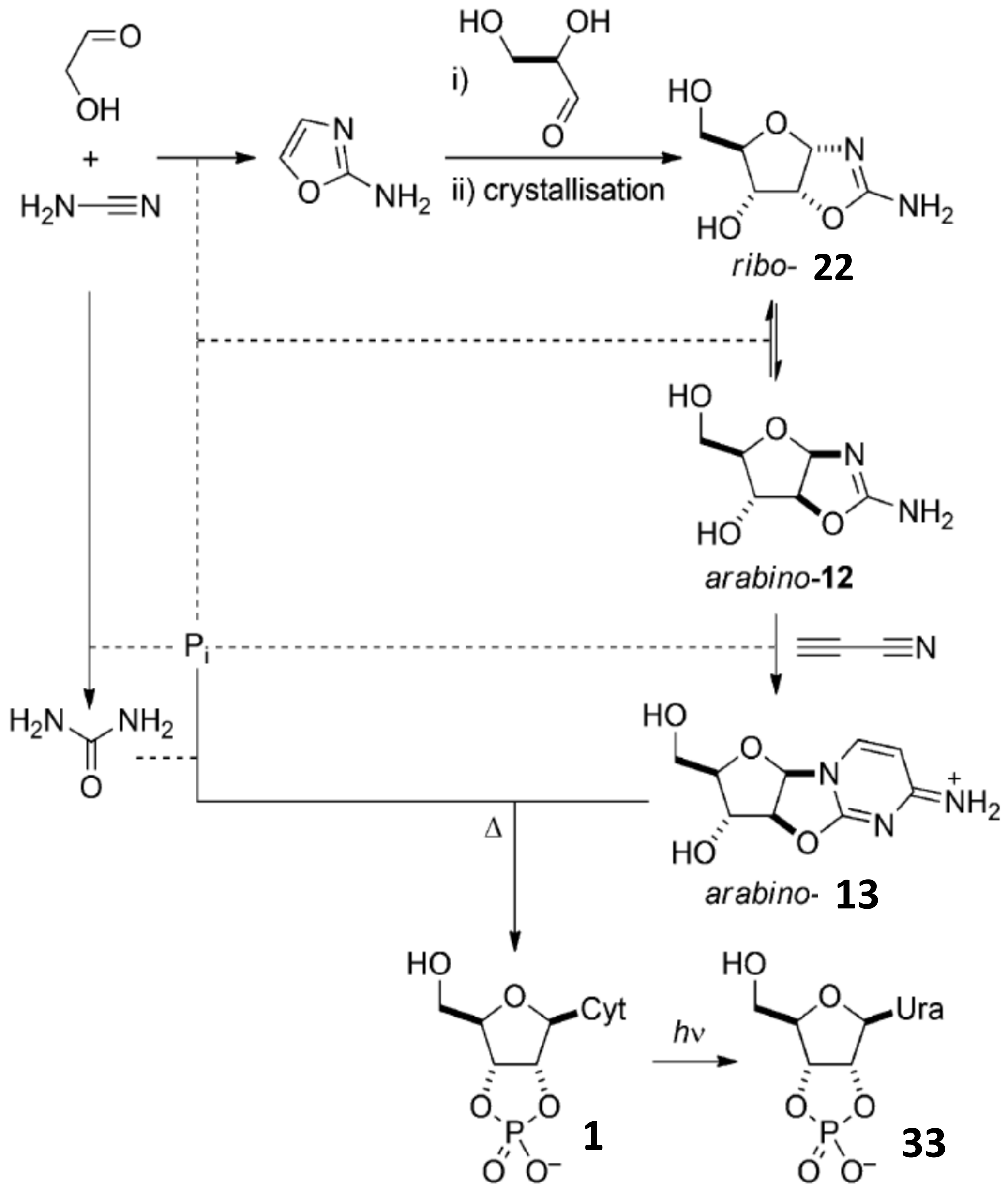
step 5: UV-light induced rearrangement C→U + cleanup of the side products



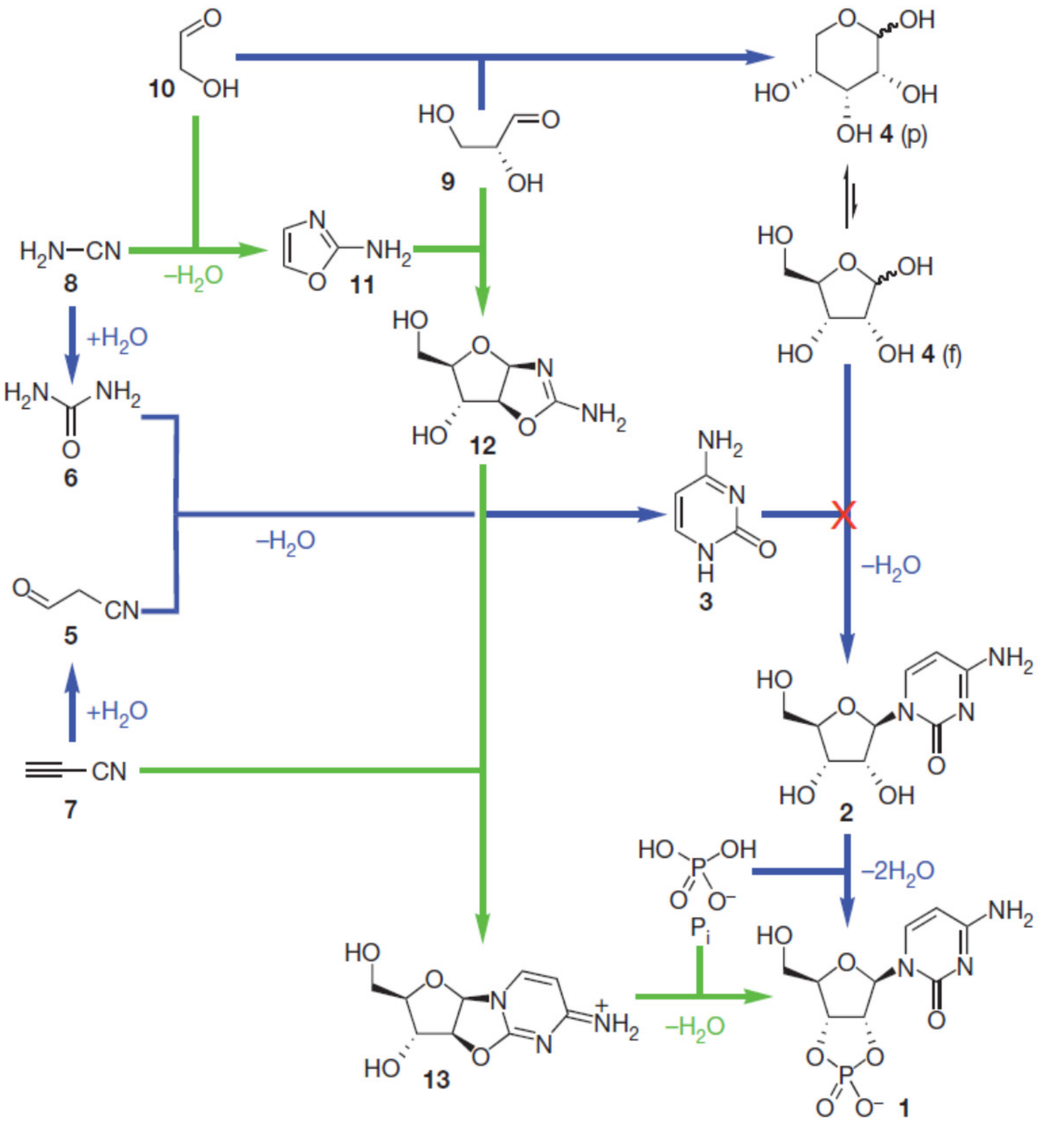
Photochemistry of *beta*-ribocytidine-2’,3’-cyclic phosphate **1**. Under conditions of irradiation that destroy most other pyrimidine nucleosides and nucleotides, **1** undergoes partial hydrolysis and slight nucleobase loss. Ura, N1-linked uracil; Cyt–H, cytosine; Ura–H, uracil.

Prebiotic synthesis of activated pyrimidine nucleotides

Catalysis, and reaction control through pH and chemical buffering, is indicated by dashed lines.



Prebiotic route to pyrimidine nucleotides



Prof. John Sutherland
Laboratory of Chemical Biology
Cambridge, UK

Dr. Matthew Powner
University College London, UK

Summary

Small biorelevant building blocks:
HCN, NC-CN, cyanamide, cyanoacetylene, ammonia, H_2S, phosphates, formaldehyde
available on the early Earth due to meteorite delivery (Late Heavy Bombardment), condensation in water,
precipitation as cyanometalic complexes and thermal metamorphosis thereof.

Systematic access to simple carbohydrates possible under uncomplicated set of conditions

Separate synthesis of carbohydrates and nucleobases plausible under prebiotic conditions. However, combination of these blocks into nucleosides unlikely

activated pyrimidine nucleotides (cyclic) can be synthesized by a combinatory approach, alternating both sugar- and nucleobase-yielding reactivity