H. Bridle

PHYSICS 214* K. Brasch

Date	Lectures and Topics for Weeks 7 & 8 (1979)
February 27.	Definition and basic elements of life on Earth - essential components of living systems.
March 1.	The origin of life on Earth - the evolutionary and fossil records
March 2.	Experiments on the origin of life and extreme limits of existence.
March 6.	Mars & Viking - The unfinished experiment?
March 8.	Other life systems - Venus, Jupiter & Titan
March 9.	Other life systems - Panspermia - General Discussion.

General References (Available in either Physics or Bio. Libraries)

S.W. Fox and K. Dose - "Molecular Evolution and the Origin of Life" - Marcel Dekker Inc. 1977.

S.L. Miller and L.E. Orgel - "The Origins of Life on the Earth" - Concepts of Modern Biology Series - Prentice Hall 1974.

C. Ponnamperuma (ed.) - "Chemical Evolution of the Giant Planets" - Academic Press 1976. *(Not in library but I have copy.)

R. Jastrow - "Report from Mars" - Natural History - March 1977.

N.H. Horowitz - "The Search for Life on Mars" - Scientific American, Nov. 1977.

M.R. Heinrich (ed.) - "Extreme Environments" - Mechanisms of Microbial Adaptation - Acad. Press 1976 (several chapters).

L. Margulis, H.O. Halvorson, J. Lewis, A.G.W. Cameron, 1977 - "Limitations of Growth of Microorganisms on Uranus, Neptune and Titan"- Icarus 30, 793-808.

N.C. Wickramasinghe, F. Hoyle, J. Brooks and G. Shaw, 1977 -"Prebioitic Polymers and Infrared Spectra of Galactic Source"- Nature 269, 674-676.

C. Ponnamperuma, 1976 - "The Organic Chemistry and Biology of the Atmosphere of the Plant Jupiter" - Icarus 20, 321-325.

W. DeWitt - 1977 - "Biology of the Cell - An evolutionary approach."
W.B. Saunders Co.

LECTURE NOTES (1979)

Physics 214

K.R. Brasch

Definition and Basic Elements of Life on Earth

The precise definition of "life" or "living matter" is a classical philosophical and scientific question. For example, plants and animals are clearly living, while rocks and water are clearly non-living or inanimate entities. In spite of these obvious considerations, however, the individual attributes and/or components of life are not clearly or unambiguously defined, eg:

1. Reproduction & Growth - these characteristics of living systems can be simulated both artificially and naturally.

   For example: artificial cells (co-acervates/microsphere), inorganic and organic crystals (eg. Si carbide), machines, etc., all can exhibit or simulate growth and reproduction.

2. Energy Consumption - i.e. Organisms require constant input of thermal/ chemical energy from external environment. However, many endothermic chemical reactions do the same, and all machines need energy to work.

3. Organized & Complex Matter - True living systems are highly organized and complex forms of matter, but so are crystals, organic cpds, natural polymers, etc.

Several additional questions are also related to a definition of life. Are dormant seeds and spores alive? Dessicated seeds $\sim$10,000 yr old have been found in neolithic graves, etc. Clearly yes, they are "alive", but in a state of suspended animation. Thus most basic criteria attributed to living systems can be partly or fully simulated or re-enacted (chemically in test tube, mechanically or by computers) - i.e. "Living" organisms in many ways are more than sum of their individual components. (NB - the antithesis "dead" simply implies "no longer alive" ∴ not same category as "inanimate" - i.e. "never having been alive".)

Philosophical questions aside, however, how can we pragmatically or operationally define "Earth" life as we know it? The following characteristics apply:

1. All organisms are composed of basic units called "cells" - the cell is the ultimate structural and reproductive entity of an organism. Cells are physically and chemically limited from the external environment and exhibit semi-permeable uptake and release of external and internal compounds through the cell boundary (membrane/wall).
2. Cells arise only from pre-existing cells - i.e. there is always continuous lineage and precise self-replication. Cells do not arise *denovo* or spontaneously (e.g. like crystals/polymers).
3. Cells are net energy (E) accumulators (sinks) - i.e. they require constant input of E from the external environment to build up and synthesize "self". Two categories of E assimilation exist:
   - A) Direct assimilators (light-activated primary analysis). These are called:

     Autotrophs (photosynthesizing bacteria, blue green algae and green plants).
   - B) Indirect E assimilators (via nutrients). These are called:

     Heterotrophs (animals, fungi, non-photosynthesizing organisms).

Following is a summary of the basic (essential) elements of living systems on Earth. The simplest or common denominators are: the structural elements of cells, their molecular aspects and thermodynamics.

I *Structural* elements of cells (Fig. 1).

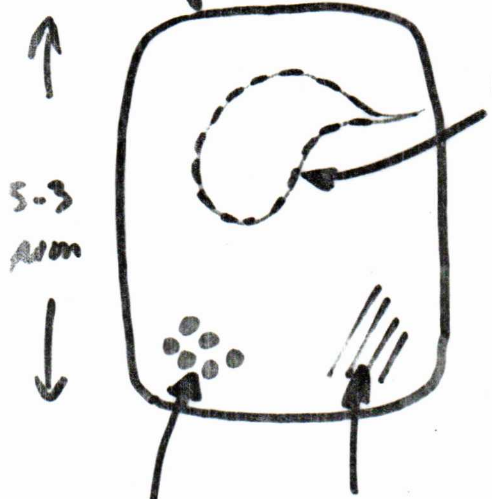
Structural Elements of Simple Cells (Prokaryotes)

① Cell Boundary (wall and membrane)

function — composition

- limiting biochem environmt.
- selective exclusion
- protection

} lipid, protein } dynamic complex
\+ carbohydrate (cellulose) rigid support



② Genetic Apparatus

coding system for storage/tranfer of info. specifying the organism } DNA (RNA)

③ Energy Assimilation Apparatus

Trapping/use of energy for biosynthesis

Photosynthesis → lipid protein + photo pigment

Bio-redoxitation → lipid protein + enzymes

④ Translation (decoding) Apparatus

protein synthesis
ie. make enzymes for **all** biosynthetic catalysis required by the cell

} RNA protein (Ribosomes and t-RNA)

II <u>Molecular Aspects of Cells</u> - The following three main considerations apply: - Elemental abundance, molecular organization, information transfer.

(a) Elemental Abundance (Fig. 2).

(b) Molecular Organization (see Ref. & Supplements).

All biological macromolecules are biopolymers and include:

1. Nucleic acids - DNA; RNA. A major attribute of this group of molecules is their limited shape, which is ideal for replication, coding and decoding.
2. Proteins. These molecules in contrast can have an infinite variety of shapes and structures, which is ideal for cell structure, enzymes, etc.
3. Carbohydrates (sugars). These molecules are easily assembled and broken down, and thus serve as main sources of chemical energy/storage.
4. Lipids. Function in both structure and energy storage/assimilation. Their hydrophobic properties aid tremendously in this respect.

(c) Information Transfer Systems (Nucleic Acids).

This is probably the single most important aspect of living systems. It is essential for two main reasons, because an info transfer system must provide both:

1. Stability and hence continuity of species. Individual cells of a species die, are consumed, mutated, etc., but the genetic code (DNA/RNA) is maintained to make more of same. Thus DNA/RNA is a bank of info bits designed to self-perpetuate.
2. Variability and hence make evolution possible. Because the genetic apparatus is also susceptible to small changes, e.g. mutations, recombination, these permit trial and error variability, hence selective adaptation and production of chains of organisms of increasing complexity.

Chemical Composition of Terrestial Life

Of all elements ~ 25 used by living organisms

Relative Abundance of Elements (atoms/100 atoms)

Element	Universe (estimate)	Earth Crust (approx.)	Sea Water (approx.)	Soft Tissue of Cells
H	90.80	2.9	66	63 *
He	9.08	t	t	—
Carbon	0.02	0.03	0.001	11 *
N	0.04	t	t	1.1 *
O	0.06	60.6	33	25 *
Na	trace	2.5	0.3	0.03
Mg	"	1.8	0.03	0.02
Silicon	"	20.6	t	t
P	"	0.07	t	0.2 *
Sulfur	"	0.02	t	0.3 *
Cl	"	t	0.3	0.03
K	"	1.4	t	0.002
Ca	"	1.9	t	0.4
Fe	"	1.8	t	t

t = trace < 0.003 * Key elements for biomolecules

- During biol. evolution certain element selected for use, others rejected (eg He)

- Reason - prob. { chemical nature of atom, reactivity, stability of cpds formed etc

There are several components or considerations involved in the evolution of info transfer in biosystems. First, a coding method must evolve which works and is stable from generation to generation (i.e. provides genetic continuity) and can be decoded within a given generation. An analogy here is a record or tape. This can be copied, and a permanent replication made, or you can play (translation/transcription) i.e. make a transient copy of the stored information (Fig.3). Only nucleic acids can fulfill this role adequately. From an evolutionary standpoint 2 genetic codes are required - a primary system (codons) and a secondary system (anticodons) plus recognition enzymes. This is not easily explained or accounted for by most theories of origin of life.

III Thermodynamics and Energy Assimilation - All biochemical energetics are fundamentally reduction/oxidation reactions - i.e. an alternate loss/gain of e's and protons (H^+) is involved. The net liberation and exchange of free E from breakage and formation of chemical bonds provides the thermodynamic basis for sythesis and degradation of all biomolecules, e.g.:

1. Carbohydrates (universal potential E sources).

$$C_6H_{12}O_6 + O_2 \longrightarrow 6CO_2 + 6H_2O \qquad \Delta E$$

(Highly reduced form of C) (Oxidized) (Very oxidized form of C) (Very reduced form of Oxygen)

2. $ADP + Pi \xrightarrow{\Delta E} ATP \longrightarrow$

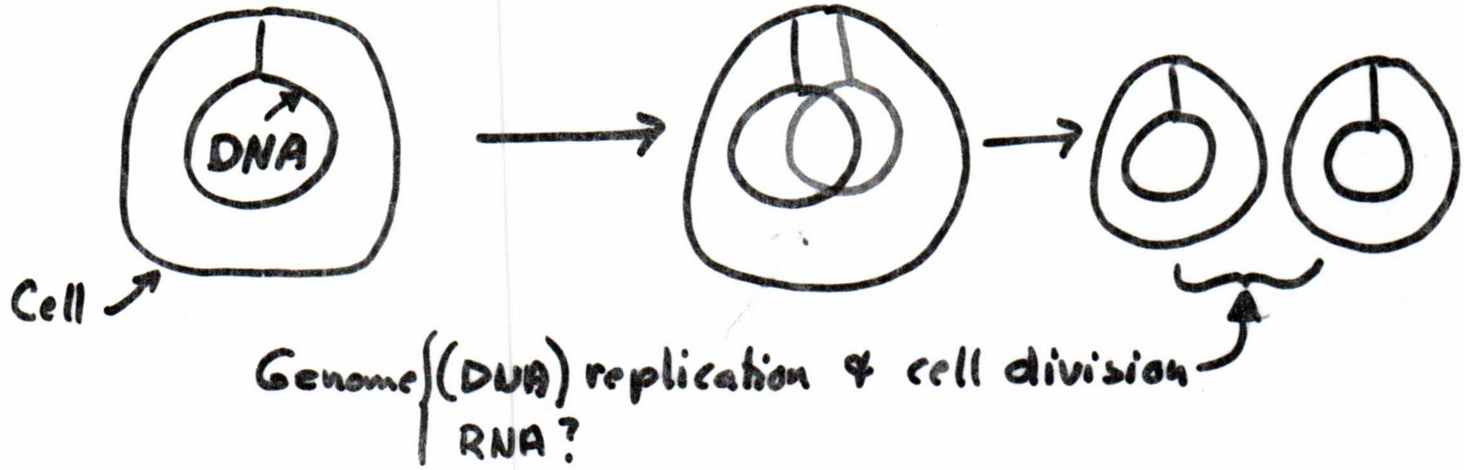
Oxidized (low potential E cpd.) Reduced (High potential E cpd.)

NB - A net external E input is essential - i.e. organisms do not contravene the 2nd law of thermodynamics by going from simple to the more complex.

igure

Essential Aspects of Info Transfer

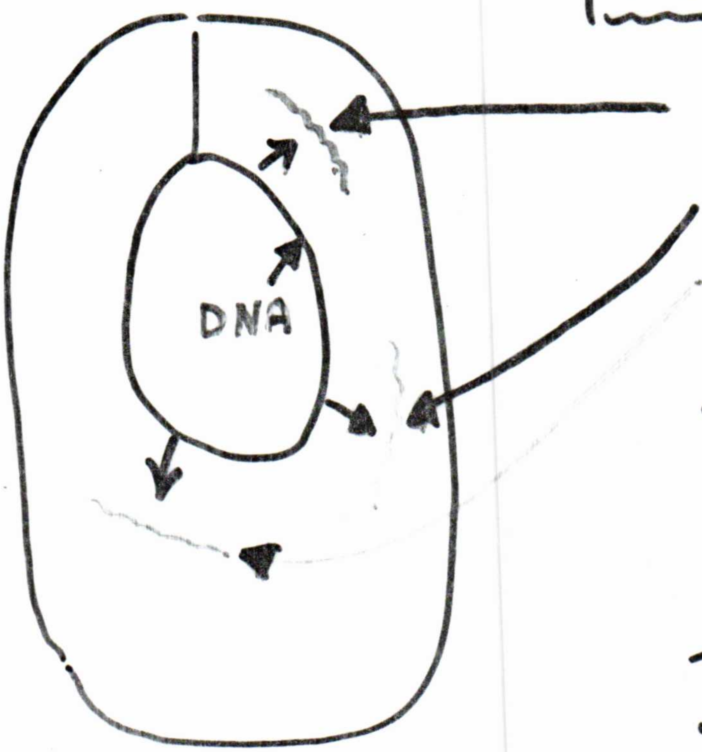
① Genetic continuity (Generation to Generation)



② Intracellular decoding (within Generation)

3 classes of genes:

① transfer RNA
② Ribosomal RNA
③ messenger RNA



Transcription (decoding)

{ Synthesis of 3 types RNA from DNA

Translation

{ Synthesis of proteins by 3 types of RNA

In this context, 3 main points are important concerning the evolution of life. Organisms may be aerobic (O_2 requiring), anaerobic (non-O_2 requiring) and/or capable of photosynthesis as primary energy assimilation (Figs. 4 & 5).

The Origin of Life on Earth (and it's effect on the planet's evolution)

To fully appreciate the major aspects of this question, the following components must be considered: (a) the geological conditions on primitive Earth; (b) the "fossil" record; and (c) the evolution of higher organisms.

(a) Geological conditions on primitive Earth.

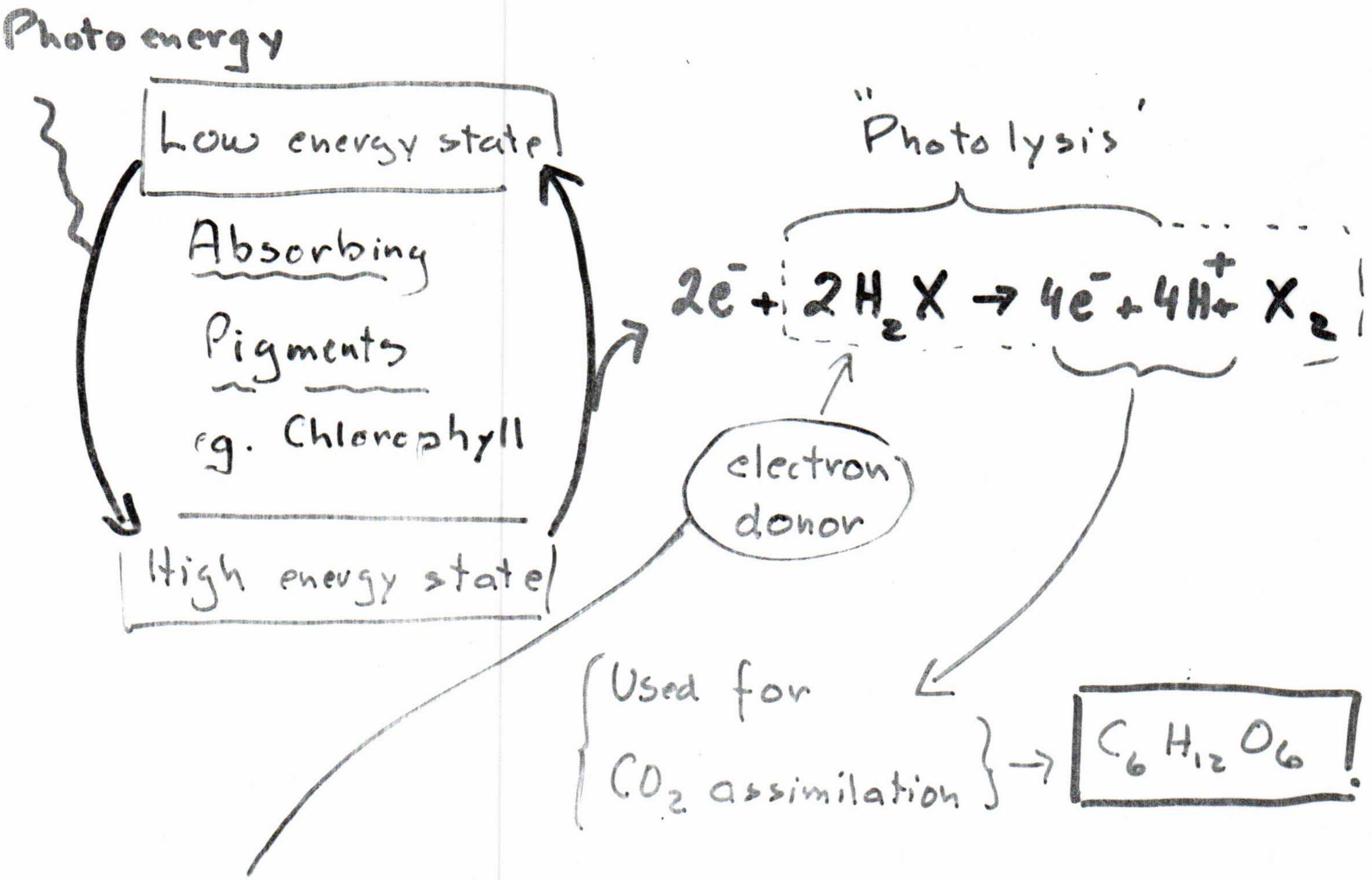
These are very difficult to assess with any certainty, because the Earth has "evolved" since its formation such that the initial conditions have been changed or obliterated by such factors as tectonic activity, weathering, life on large scale, etc. In terms of life related aspects, however, the following picture emerges and must be explained: (Fig. 6). A particularly difficult problem is to picture the very young Earth under "pre-biotic" conditions, since such conditions presumably were either limiting or conducive to emergence of life. Thus, an understanding of pre-biotic geology is crucial:

In terms of the lithosphere - consisting of a core, mantle, and crust - least is known about the latter and yet it is very important re life, because the crust must: (a) provide a semi-solid surface for condensation/concentration of various organic molecules; and (b) furnish essential metallic elements - Ca, Mg, Na, K, Fe, etc. for enzymatic reactions, binding, cell walls, etc.

The hydrosphere is clearly important in terms of life, since the presence of liquid H_2O is crucial, particularly at a reasonable temperature

Primary Energy Assimilation

Photosynthesis



$$2e^- + 2H_2X \rightarrow 4e^- + 4H^+ + X_2$$

If primary e^- donor is $H_2O \rightarrow 4e^- + 4H^+ + O_2\uparrow$

Blue green algae and all plants

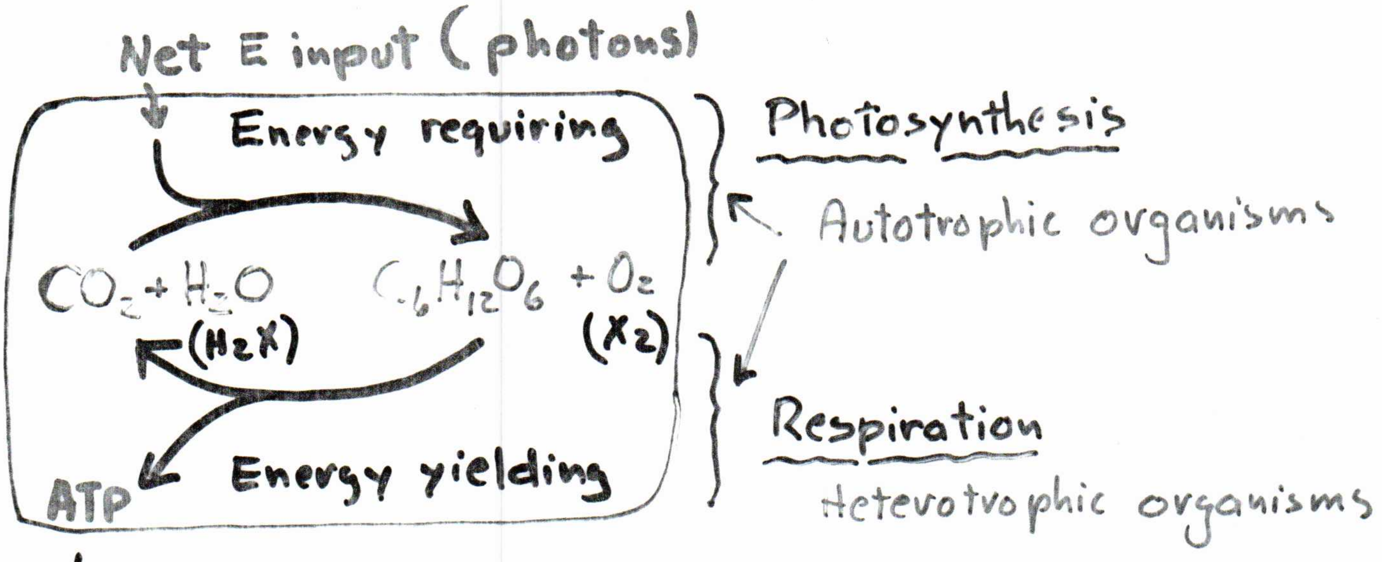
liberated into atmosphere

However, many other compounds will do:

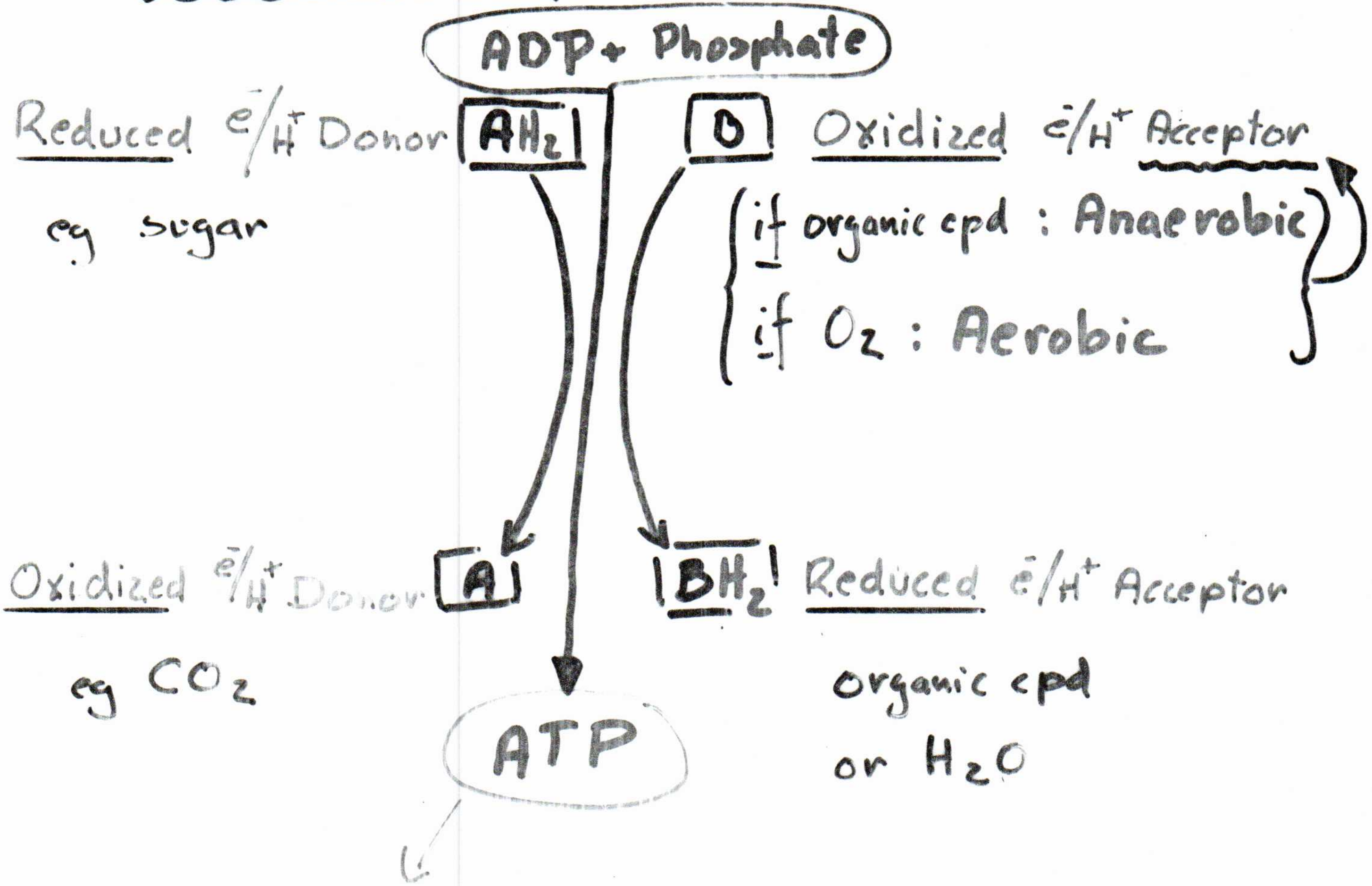
H_2X (H_2S, H_2, etc)

Photosynthic bacteria and probably all ancestral organisms

Summary of Primary Energy Relations in Cells



Generation of ATP



"Drives" most other biochemical reactions in the Cell

Figure ⑥

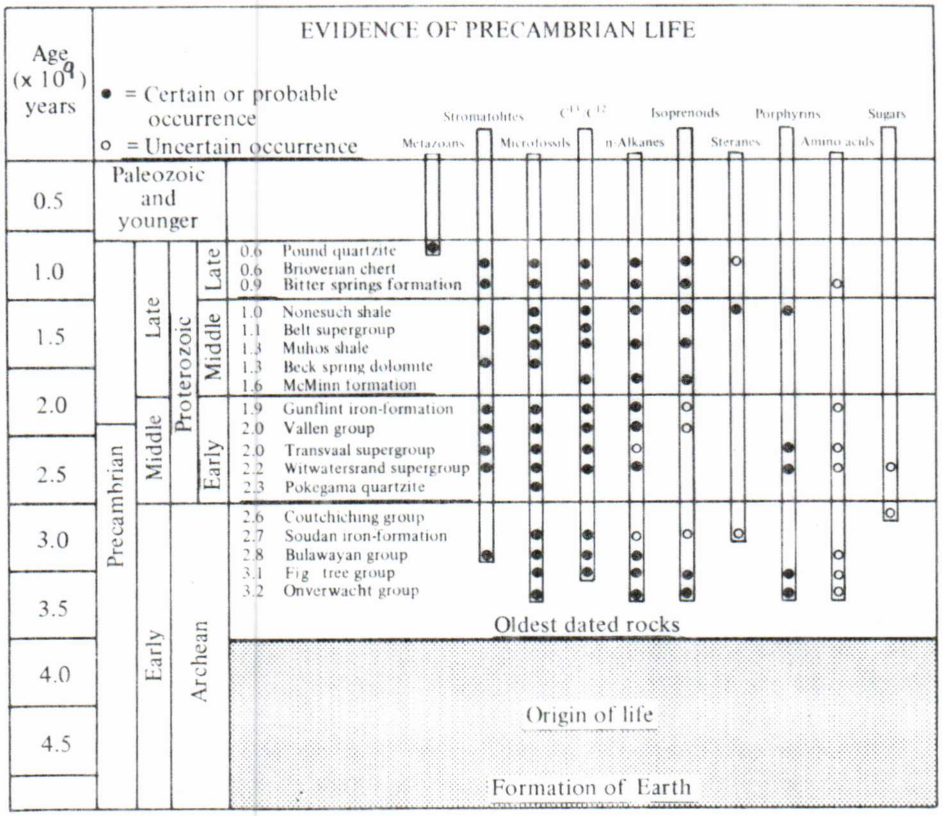


Fig. 3-5. Histogram showing the distribution of organic and morphological evidence of organisms in Precambrian sediments. (Adapted from Schopf, *Biological Reviews* **45**, 323, 1970.)

and pH range. The exact time and mode of ocean formation is unclear, as is the formation and action of clouds, evaporation, precipitation, etc. A liquid, solid, gas interface is envisaged for organic synthesis, concentration and polymerization.

The atmosphere is probably the most important component relating to life, both is primeval and today's Earth. The atmosphere modulates all major aspects affecting living organisms, i.e. temperature, humidity, incoming radiation, etc. In addition, the availability of O_2, CO_2 is crucial as essential primary metabolites. While the exact evolution and composition of the primitive atmosphere remains uncertain, general consensus exists on the following: (a) the original "primary" atmosphere was lost during or shortly after Earth formation; (b) a "secondary" atmosphere resulted from outgassing of the Earth's interior; and (c) the modern atmosphere is totally different from original, in that it is unusually high in O_2 content (~20%), low in CO_2 and in a state of non-equilibrium.

Several models of atmospheric evolution have been proposed. All are speculative and based on different initial assumptions concerning original gas mixes, rates of interactions, temperatures, etc. Most models, however, agree that the primitive atmosphere was non-oxidizing to moderately reducing. There are varied estimates of initial gas mixes:

1. CH_4, NH_3, H_2 - highly reducing; or
2. CO_2, N_2, H_2O - moderately reducing.

The latter is favoured but all agree that no free O_2 was present. It is thought that most O_2 was tied up on oxides ($FeSO_4$, Fe_3O_4, UO_2, $CaCO_3$). Free O_2 probably evolved relatively late and/or rather suddenly. Since most atmospheric O_2 is probably due to photosynthesis, this mechanism was not present in early Earth.

The foregoing general scenario is essential to explain the origin of life because most organic compounds are unstable in the presence of free O_2, and no prebiotic synthesis of complex molecules would be possible. Presumably all early metabolism was anaerobic. Several models of the time course of atmospheric O_2 evolution exist (Fig 7).

Recently M.H. Hart has constructed several computer simulation models of the evolution of the atmosphere. This represents the most comprehensive attempt to date. Hart's model included consideration of many factors, including: solar radiation; degassing from the Earth's interior; the mean composition of initial gas mixes; the condensation of H_2O into oceans; the photodissociation of H_2O; albedo; cloud cover; and the presence of life and photosynthesis. All above factors were assigned reasonable known values and processings (i.e. no extraordinary or catastrophic events are included or needed). Several computer runs were made and best fit results in line with currently observed data were sought. When the assumed mean composition of juvenile volatiles were as follows: $\sim$85% H_2O; $\sim$14% CO_2; $\sim$1% CH_2; $\sim$0.2% N_2, the mix would lead after 4 billion years (BY) to the closest approximation of present day conditions of gas mixtures, temperature (mean surface), and freedom from extensive cloud cover, etc. The model also fits extremely well with most expected prerequisites for conditions for evolution of life on Earth (Fig. 8).

A controversial factor is ozone (O_3). This is produced by photodissociation of $H_2O \rightarrow 2H_2+O_2 \rightarrow O+O \rightarrow O+O_2 \rightarrow O_3$. Ozone absorbs short λ UV light. This is good because it prevents the photodissociation of complex organics and radiation damage or mutations to organisms. The presence of O_3, however, also removes a potential E source for prebiotic

Models of time course of O_2 in the atmosphere

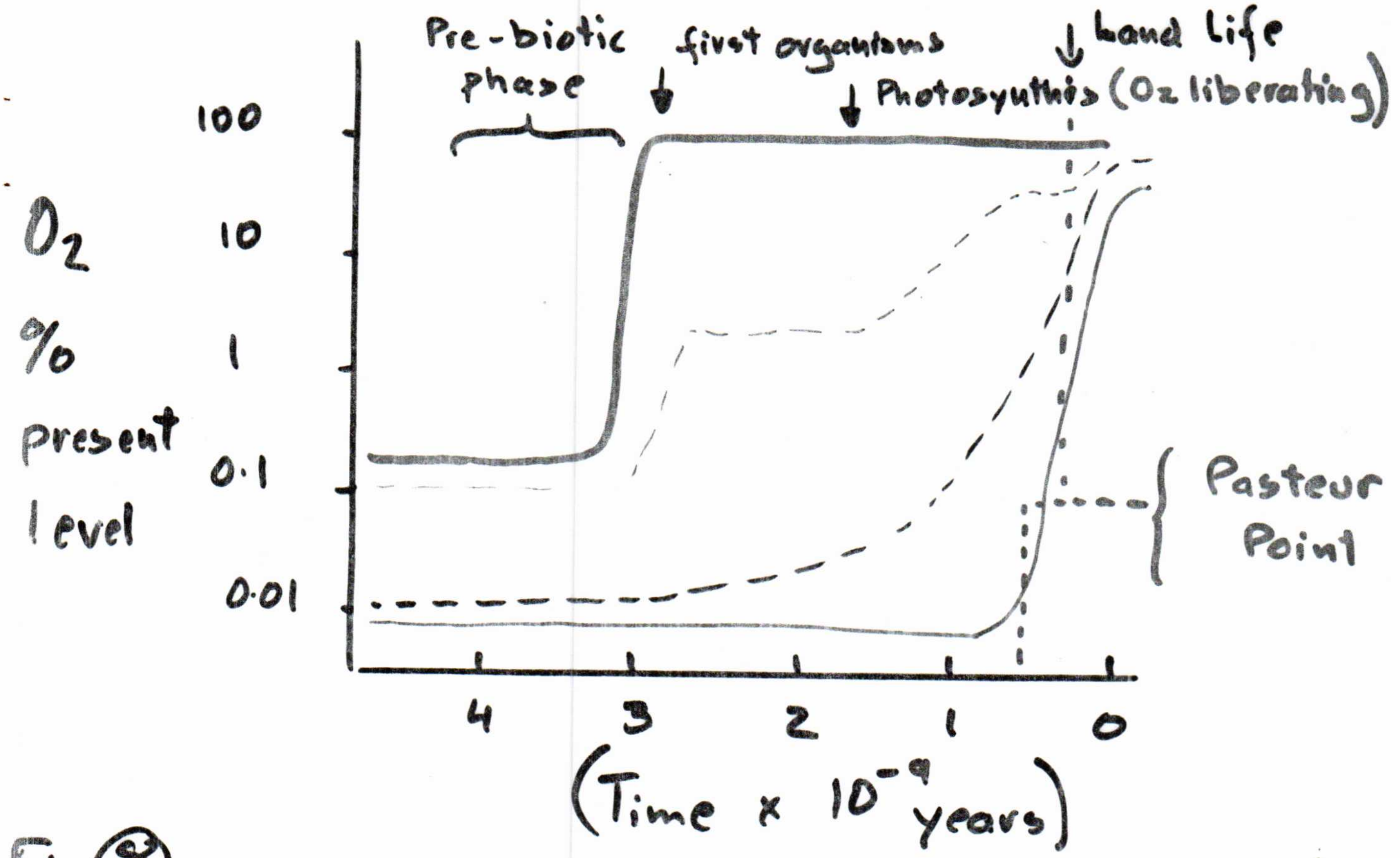
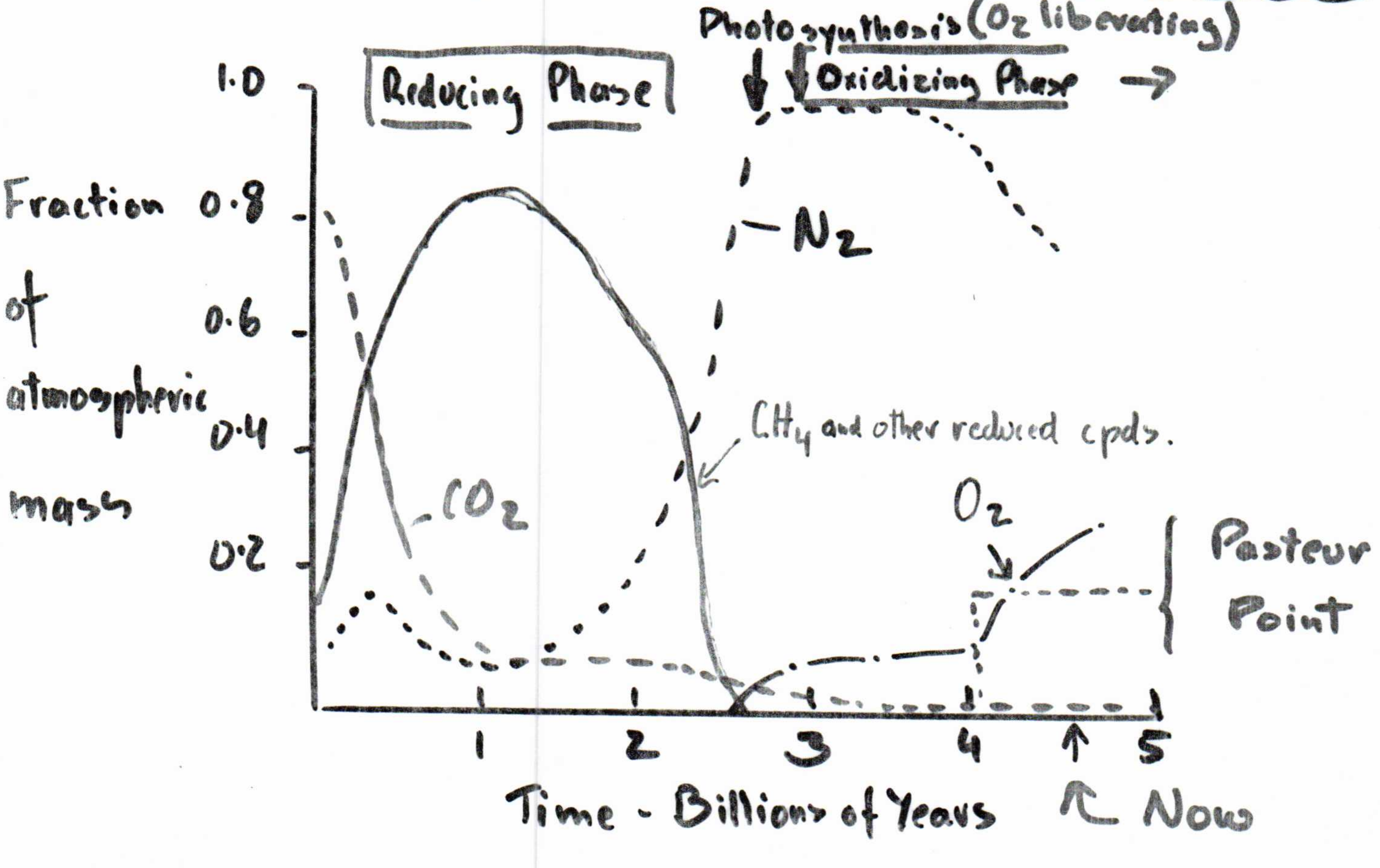


Fig. ⑧

Hart's - Computer Simulation - Best fit model



synthesis and also depends on the presence of O_2, which itself is problematic. The Hart model predicts enough O_3 present in upper atmosphere about 0.5 BY ago. This is co-incident with the spread of life to continents from oceans and if true, may imply that UV radiation was too intense before to permit higher evolution on land.

(b) The "fossil" record of life.

The term "fossil" is used here in a very loose sense indeed and includes any fossil evidence relating to the evolution of life, such as chemical deposits (both inorganic and organic), structural and metabolic fossils, etc. How does the record fit in with models of the geological evolution of the Earth? Organic compounds (e.g. amino acids, fatty acids, sugars, etc.) are present in many ancient rock deposits ~~up to~~ 3.5 BY (+) old. This is not sufficient evidence of life, however, since this could simply represent contamination from more recent organisms. Stromatolites and microfossils are also found in many ancient rock deposits. Stromatolites are limestone deposits in stratified layers of $CaCO_3$ plus microbial debris. Sediments of this nature are deposited today by microbial colonies of algae and bacteria. However, such deposits are also produced by non-biological reactions due to ppt. of Ca^{+2} and CO_2. A major question then is, how reliable as indicators of the presence of organisms are the very old stromatolites containing "microfossils"? Lynn Margulis has done extensive research in present day salt marshes where deposits of similar morphology/organic content are formed. Identical ~~Same~~ patterns of microbes are observed in very old (3 BY), newer (1 BY) and present day deposits of this nature, indicating ancient stromatolites are true evidence of life.

What about photosynthetic fossils? Modern photosynthetic organism prefer $^{12}CO_2$ over $^{13}CO_2$ assimilation. ∴ if organic/fossilized deposits are relatively enriched in ^{12}C compared to $^{12}C/C^{13}$ ratios of non-biological sources in the atmosphere, hydrosphere and carbonate rocks, then this is considered good evidence of photosynthesis. Also porphyrins, part of chlorophyll complex in many organisms, if these are richer in ^{12}C in fossil deposits, then this is very good evidence of photosynthesis. ([redacted]). The evolution of higher organisms (Eukaryotes) can best be summarized as shown in Fig. 9 . The picture suggests that Prokaryotes emerged first (3.5 BY ago) and Eukaryotes about 1 BY ago, coincident with massive O_2 production. This eventually permitted multi-celled organisms to evolve and later the ascent of life from water to land.

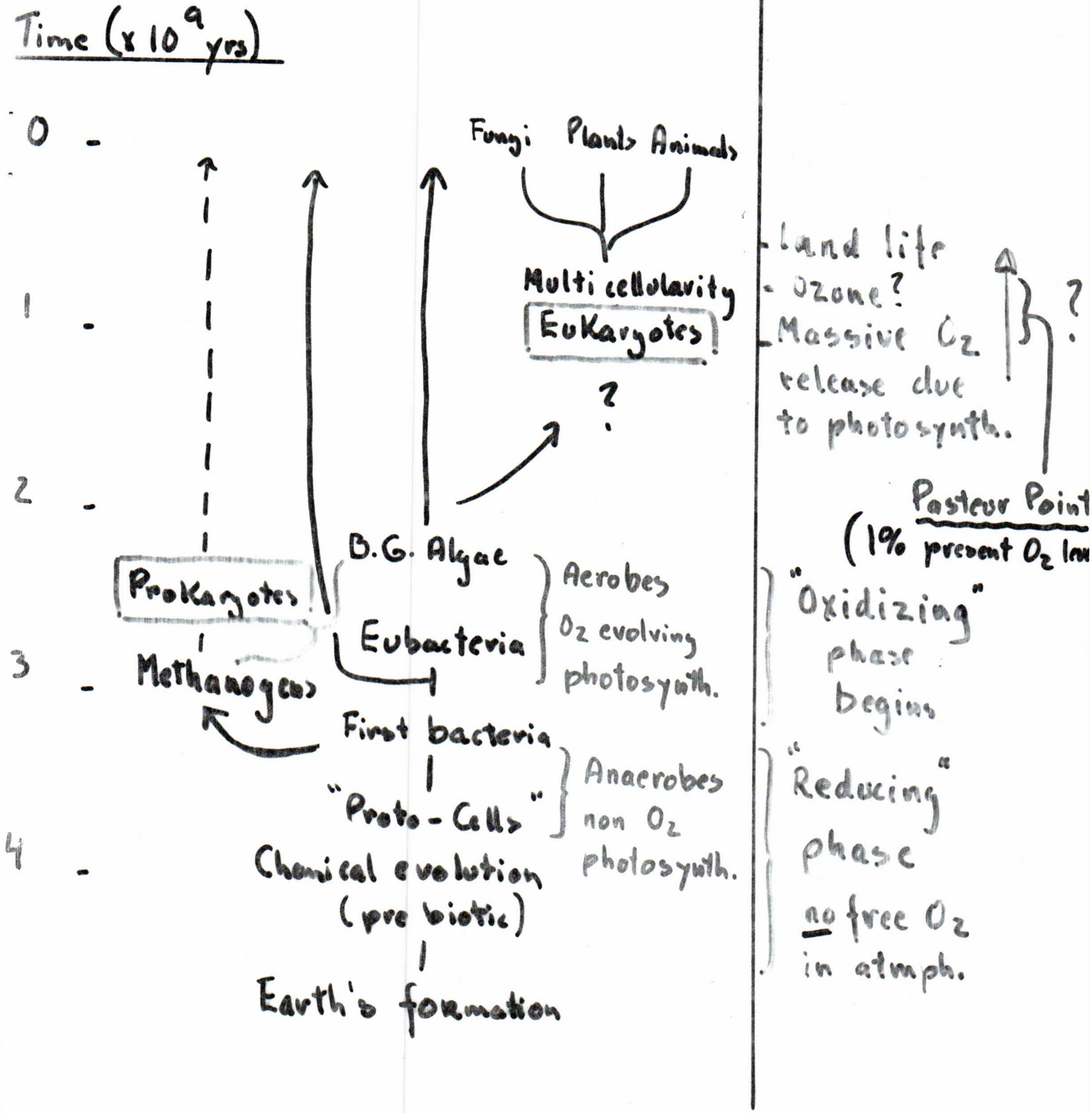
Experiments on the Origin of Life

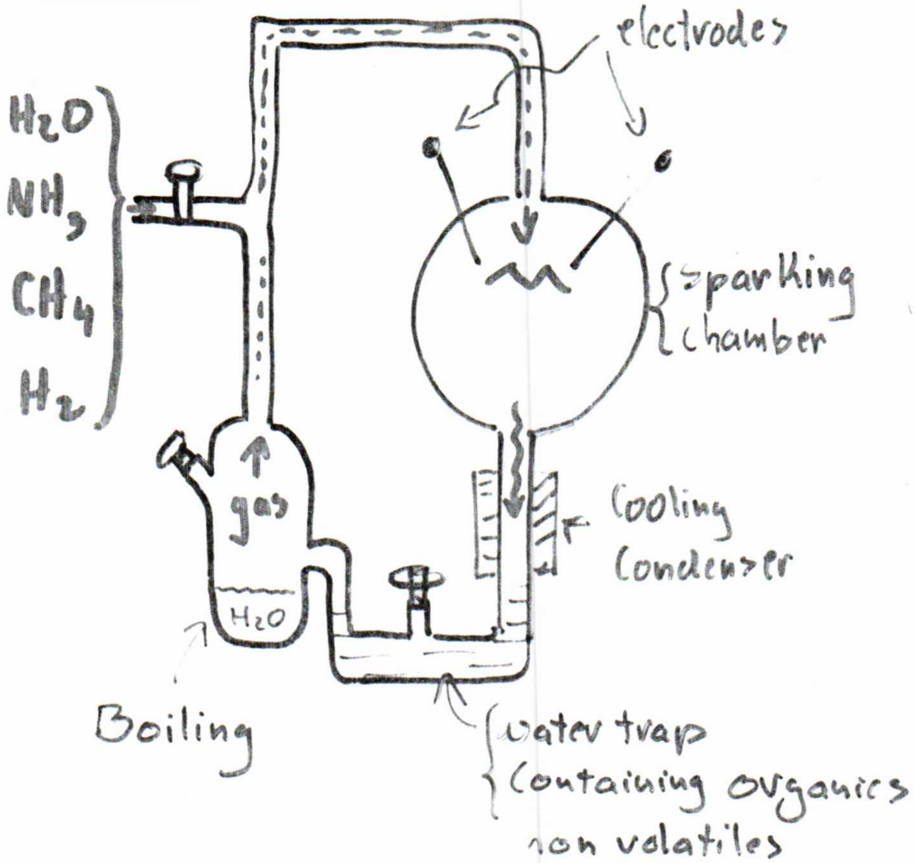
Serious attempts to explain organismal evolution in terms of the following transition:

chemical evol. → prebiotic → biochemical
(abiotic) (proto-cells) (cells)
(micromolecules) (macromolecules) (enzymatic)

were first postulated in the 1920's by Oparin, Haldane, Herrera, etc. The first solid experimental basis for non-biological synthesis of micromolecules (amino acids) - from simple inorganics, however, was provided in the 1950's by Miller. The details of these experiments illustrate hundreds of similar variations of synthesis of both micro and macromolecules from a simple mix of volatile gases in a sterile, closed system with external energy input. NB This always requires non-oxidizing conditions (Fig. 10, 10b).

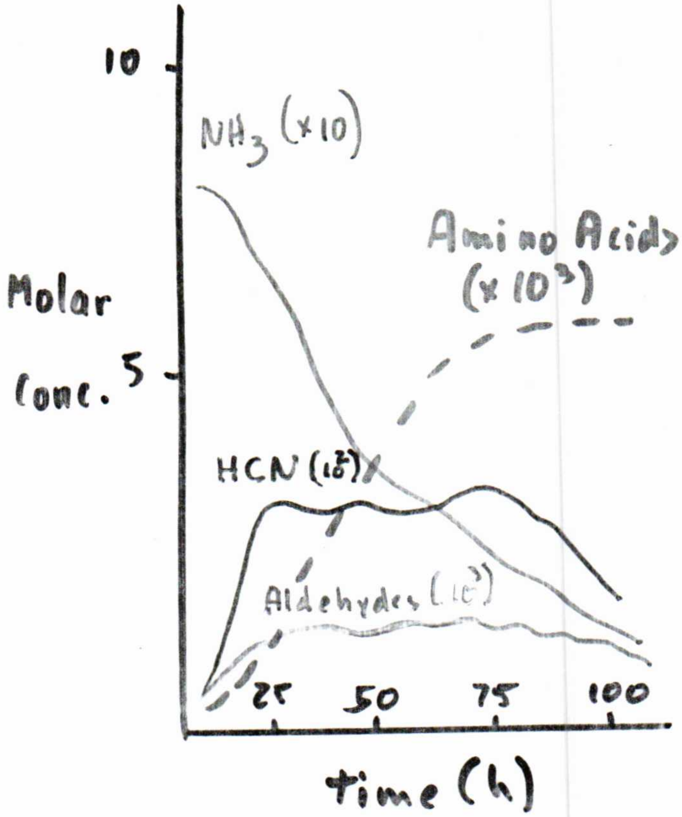
The next step of course, is how were macromolecules synthesized, i.e. the nucleic acids, proteins, triglycerides, and other large polymers of biological use? The main problem here is that on the primitive Earth, the same forces (e.g. UV, heat, etc.) required for the synthesis of large molecules is equally capable of destroying the chemical bonds forming the molecules. Thus, unless concentration and shielding of newly synthesized polymers is possible, the system soon reaches equilibrium conditions: i.e. monomer ⇌ polymer. However,

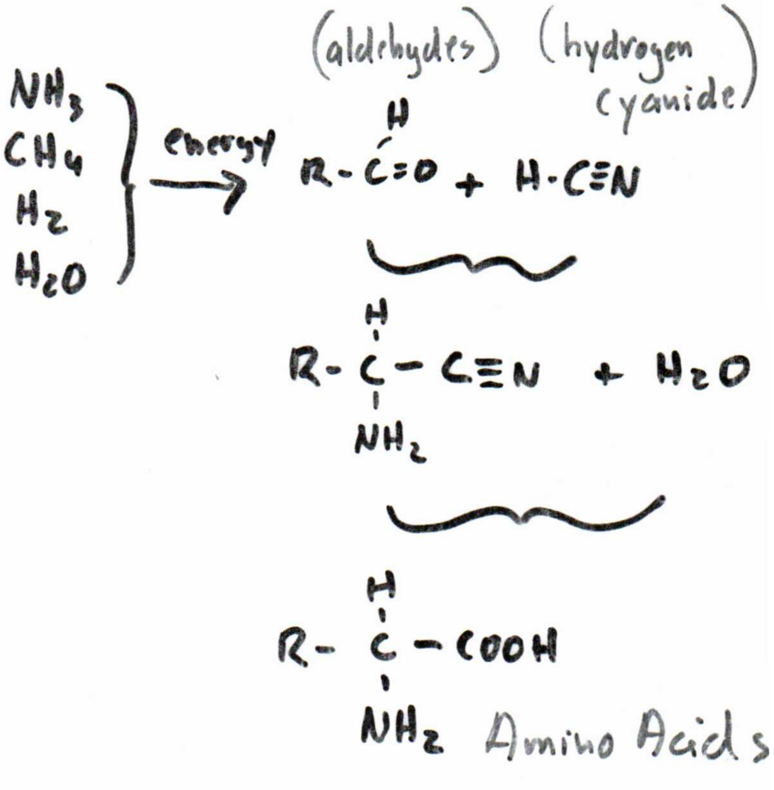

Time (x 10^9 yrs)
0
1
2
3
4
Fungi
Plants
Animals
Multicellularity
Eukaryotes
?
Prokaryotes
B.G. Algae
Eubacteria
Aerobes
O2 evolving
photosynth.
Methanogens
First bacteria
"Proto-Cells"
Anaerobes
non O2
photosynth.
Chemical evolution
(pre biotic)
Earth's formation
land life
ozone?
Massive O2
release due
to photosynth.
?
Pasteur Point
(1% present O2 lev
"Oxidizing"
phase
begins
"Reducing"
phase
no free O2
in atmph.



Various gas mixes tried which produce simple amino acids:

CO_2, N_2, H_2, H_2O,
CH_4, NH_3,
CH_4, CO, NH_3





$$\left.\begin{matrix}NH_3\\CH_4\\H_2\\H_2O\end{matrix}\right\}\xrightarrow{\text{energy}} \underbrace{R-\overset{H}{C}=O + H-C\equiv N}$$

$$\underbrace{R-\underset{NH_2}{\overset{H}{C}}-C\equiv N + H_2O}$$

$$R-\underset{NH_2}{\overset{H}{C}}-COOH$$

Figure 10 b

Purines & Pyrimidines

① $HCN + NH_4OH \xrightarrow{Heat} \text{Adenine}$

② $\text{Adenine} + \text{ribose} + \text{phosphoric acid} \overset{U.V.}{\rightleftharpoons} ATP$

③ $HCN + H_2O \xrightarrow{U.V.} \text{Guanine}$

Sugars

Formaldehyde in basic solutions $\left\{ \begin{array}{l} \text{all} \\ \text{varieties} \\ \text{of} \\ \text{Sugars} \end{array} \right.$

Fatty acids etc.

Most synthetic Reactions possible in:

{ gas/H_2O/solid micro environments
in *non* oxidizing mix
+ heat/UV/electric discharge, etc. as E souc

∴ similar to presumptive primitive Earth the following scenario is possible: hydration ↔ dehydration; high temp ↔ low temp (Fig. 11). Thus, despite apparent difficulties, microenvironments on the primitive Earth probably provided the variable conditions needed - e.g. rain puddles, cracks, aerosols, etc. for abiosynthetic reactions. Remember too, that the primitive Earth was completely sterile (i.e. no existing bugs) ∴ aggregates of macromolecules were not degraded, provided they were shielded from e.g. UV light.

The biggest and least clear question is how do we get from molecules to cells? Clearly, three main elements must form in this connection:

1. Cells ("proto" cell), i.e. a boundary for a unique internal, microenvironment and trapping of molecules.
2. A means of energy assimilation and rudimentary metabolism.
3. An information transfer system (genetic apparatus).

None of the above are easy to visualize independently, and together as unified interacting system, is even more difficult to postulate. In terms of the origin of cells, the following experiments have been carried out, producing microscopic structures called "microspheres" and "co-acervates" (Fig. 12).

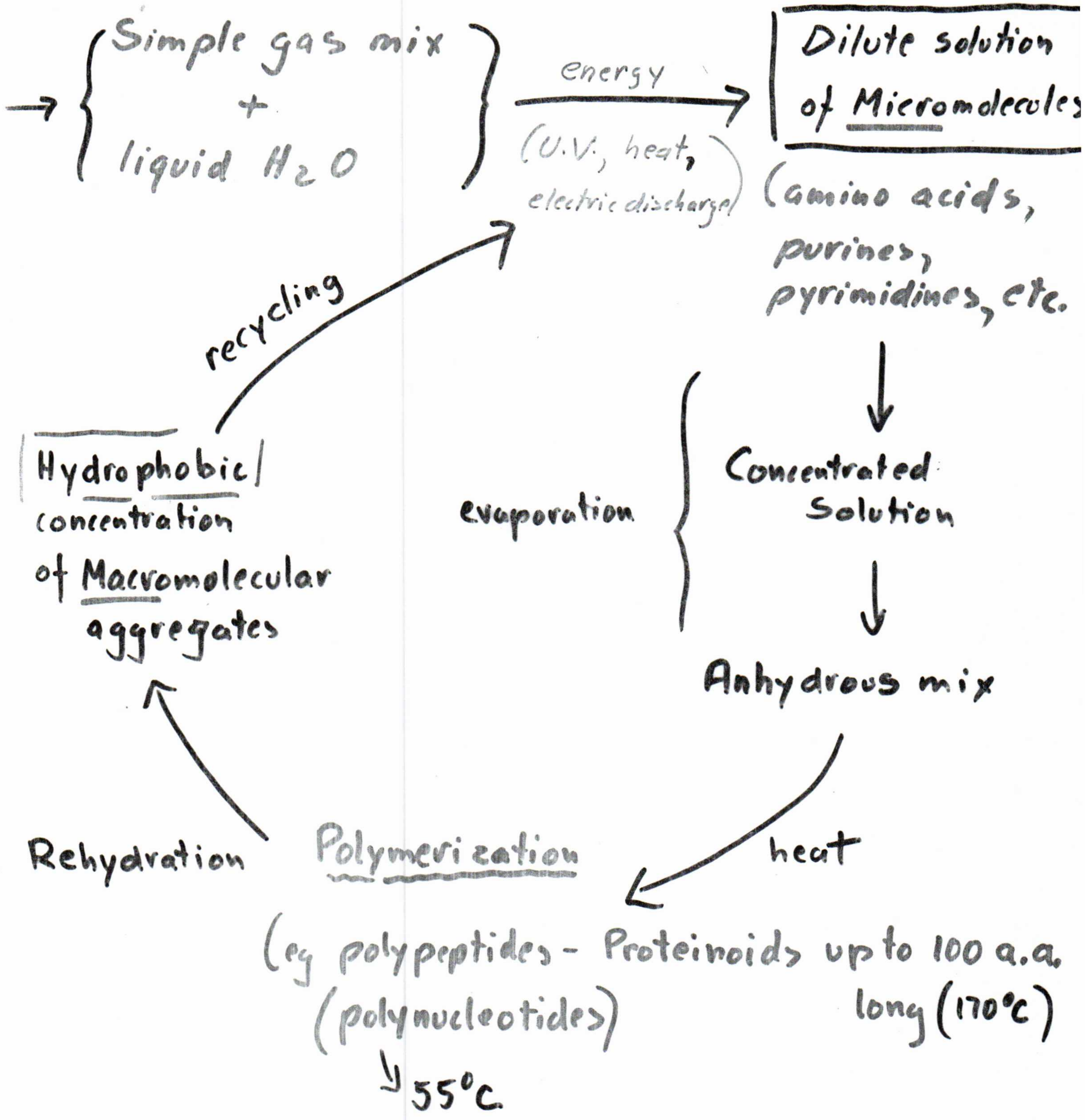
Energy assimilation and basic metabolism can be seen in terms of two factors primarily:

1. the original synthesis and condensation of micro and macromolecules, energized by light, heat and electric discharge.
2. primitive catalytic activity, i.e. proteinoid concentration into microspheres tends to favour hydrolysis of ATP to ADP for example, and mimic enzymatic activity.

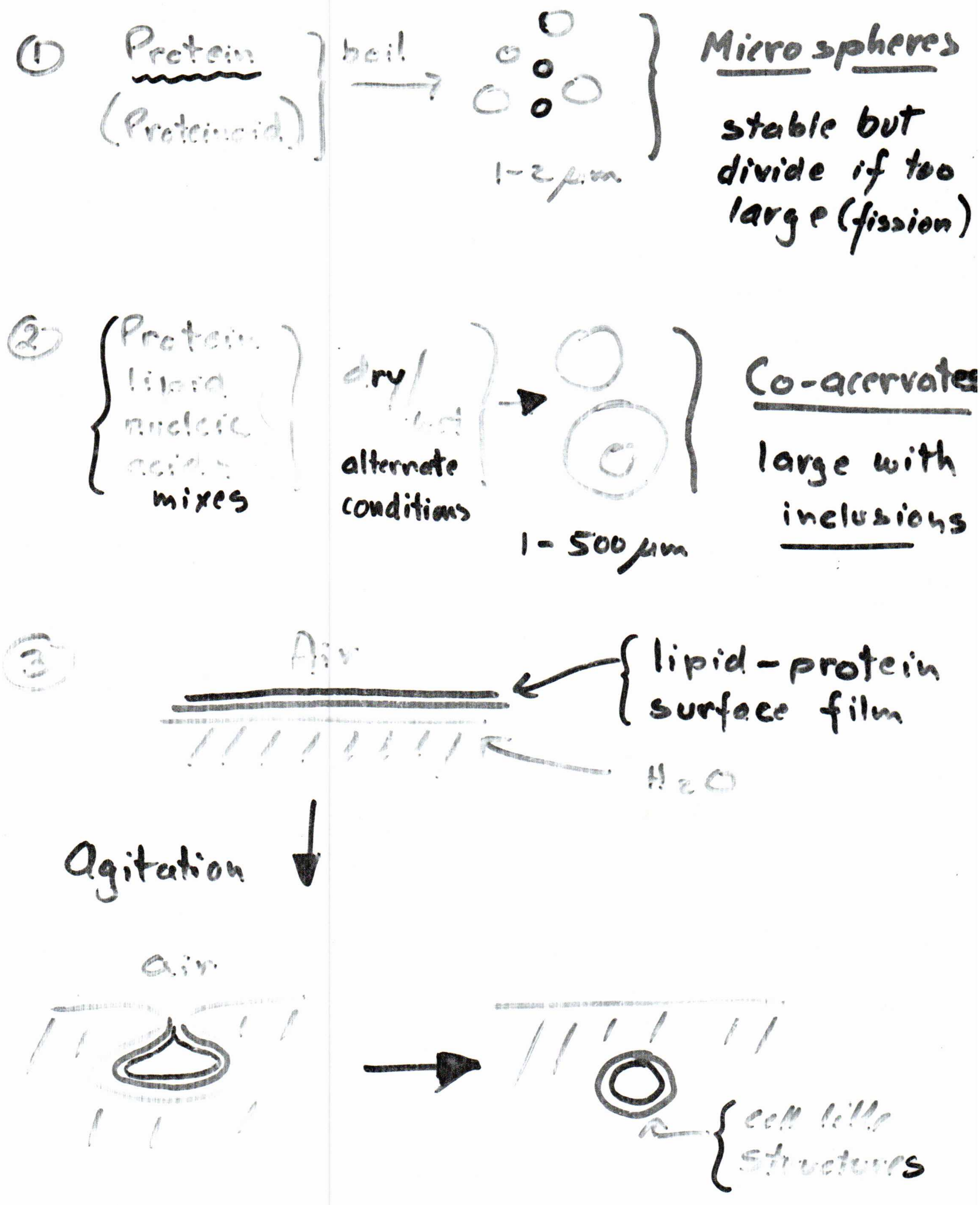
Co-acervates tend to mimic much more complex metabolic steps (Fig. 13).

In terms of the evolution of information transfer and genetic systems, this is very difficult indeed to envisage. Potentially both proteins and

Postulated Route of Macromolecule Synthesis under Primitive Earth Conditions



ig 12/ Experimental formation of Proto-cells

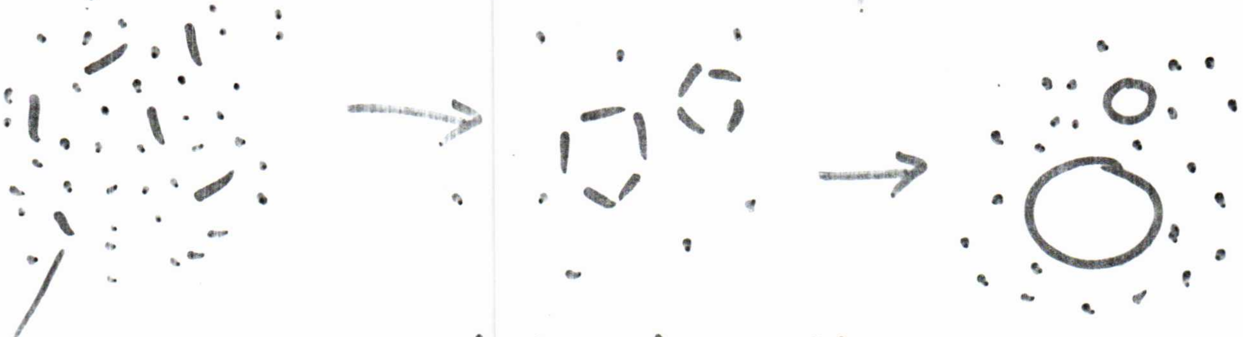


stable but divide if too large (fission)

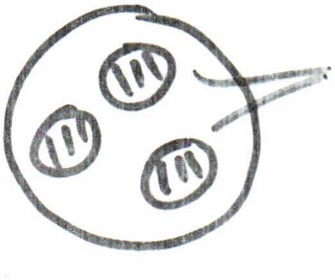
large with inclusions

Co-acervates & their properties (Fig. 13)

① Simple Coacervates : {aggregates of similar macromolecul



② Complex Coacervates : {mix of different macromolecules of opposite charge

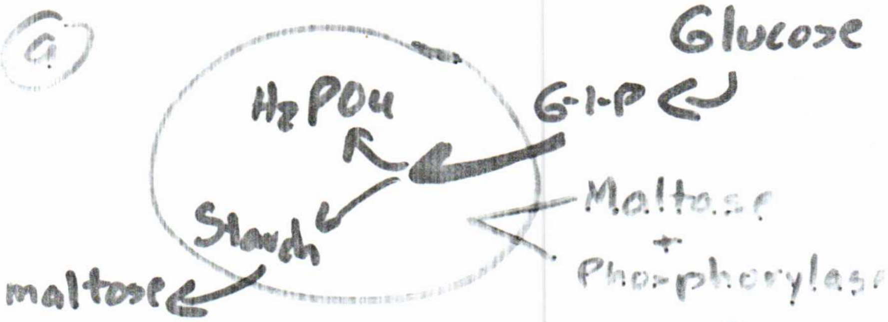


eg protein ($\pm$)
NA ($-$) (nucleic acid)
carbohydr ($-$)

③ Catalytic Properties

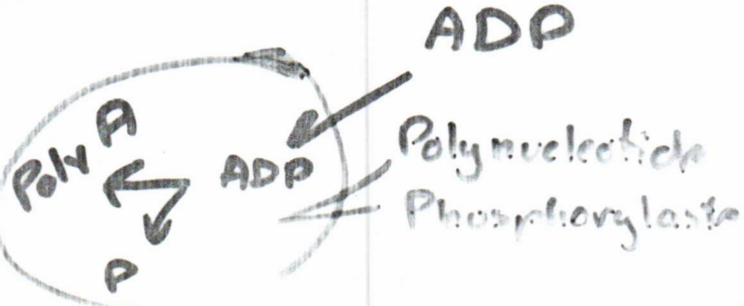
Coacervate of:

(a)



{Gum arabic (polysacchar
histone (+) protein
+ enzymes

(b)



{RNA
histone protein
enzyme

nucleic acids could function in information storage (coding) since there is enough info bit capability in both types of molecules. Clearly, however, the nucleic acids are better candidates for most of the reasons previously mentioned, including self-replication (built in) and decoding possibilities.

The main problem is establishing links between : DNA ↔ RNA ↔ proteins .
(code)(decode) translation
product

One possible model is that of Black (Fig. 14). The question remains unsolved essentially, however, since no model provides explanations for all components of genetic systems, i.e. codons, anticodons, and enzyme recognition, etc. This is particularly important since all organisms share the same info systems and no intermediates are found.

A second major difficulty facing experiments and theories of life is the macromolecule → cell transition. What was the driving force(s) behind these processes? Black suggests that the main forces were two-fold:

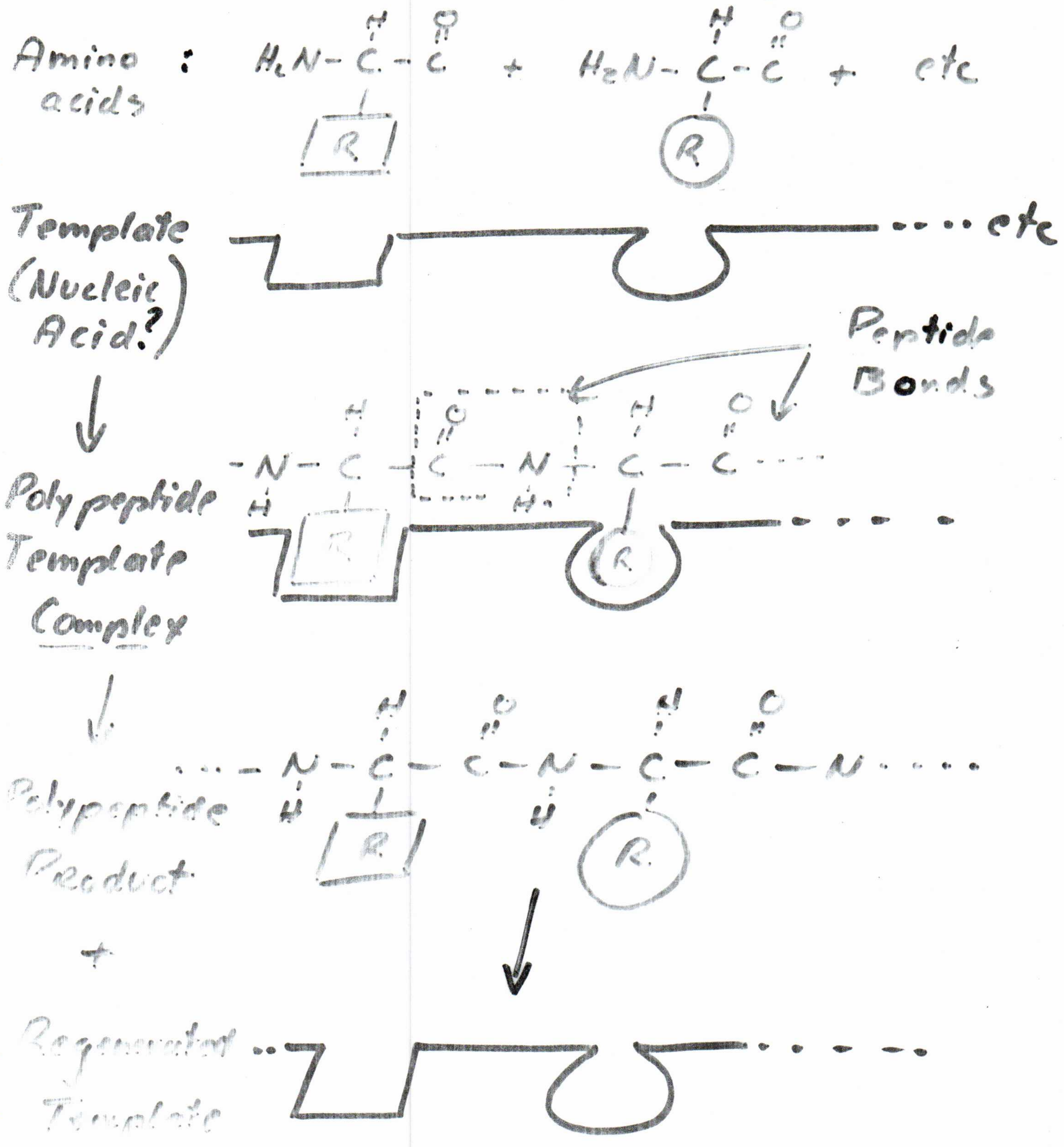
1. hydrophobic tendency (initially) of macromolecules to aggregate and exclude water; and later
2. enzymatic (catalytic) activity arose once protocells had arisen (Fig. 15).

Clearly, however, much work needs to be done in these areas, before a fully reasonable explanation is possible.

Planets and Life

To examine evolution in its broadest possible perspective we can ask questions as to whether life is unique to Earth or whether it is a common phenomenon of star and planetary system formation. Clearly the implications, possibilities, probabilities applying to this are largely unknown, but must be assessed if we are to obtain a better concept of the significance of Man's

9.4) Possible Evolution of Coding Systems

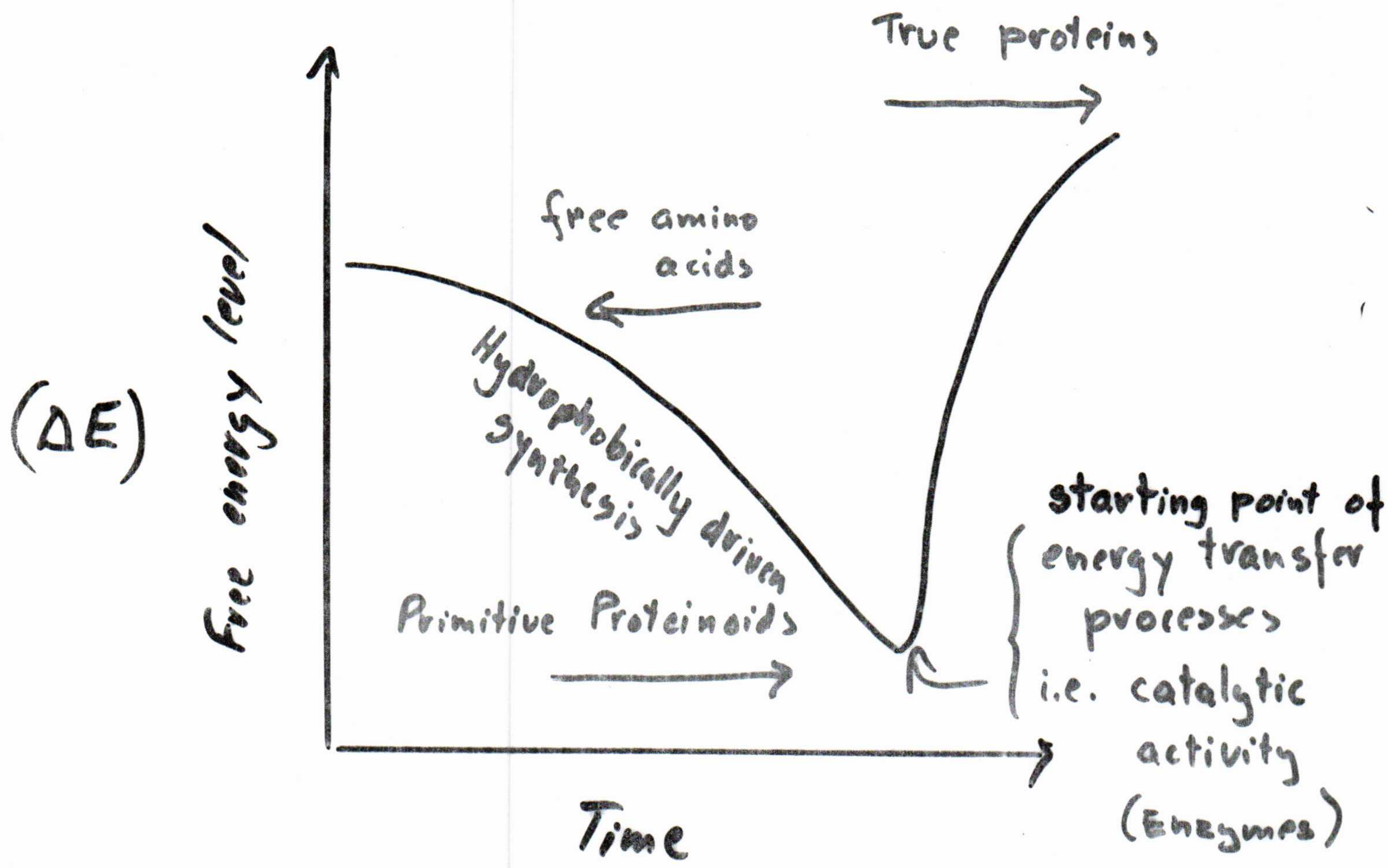


Above - method of coding is direct

Modern system is indirect ie via RNA

Figure

S. Black's - Time course model of evolution of energy phases in synthesis of proteins



place in the Universe, and the universality of life/genetic systems in general. For example, if we find life on Mars or Titan with similar biochemistry and genetic systems as life on Earth, this implies a continuity of organisms between the two planets. If the putative Martians are different, separate origins are indicated. Another important point is the time perspective. Planets take a relatively long time to form and stabilize and are probably a by-product (minor or major?) of star formation. We assume that life on Earth took a relatively long time to evolve and stabilize, yet the oldest evidence of life suggests it was present 3.7-3.5 BY ago, or about 1 BY after the assumed origin of the Earth ~4.5 BY ago. This indicates that life may have arisen much "faster" than we suspect and provides another constraint on theories of the origin of life on Earth. Clearly the subject matter is extensive and fraut with uncertainty and many "questimates". Let us examine briefly the following relevant features in this connection: the boundary conditions for life (i.e. essentials and extreme limits); Mars - as model relative to Earth and the problems raised; other life systems and alternate biochemistries; and alternate origins altogether.

Boundary Conditions and Limits of Earth Life

This subject is of interest in itself and in relation to the survival of Earth organisms elsewhere. It must be clearly defined therefore. Most organisms we know grow best within defined ranges of environmental factors (i.e. temp, pH, humidity, gas, etc.). Selection pressure presumably forces adaptations to extreme ranges and environments.

Micro-organisms are especially adaptive and interesting in this respect, because they are small (0.5-5 μm), therefore their energy/food requirements are minimal. Microbes generally have rapid generation times, allowing them to take advantage of possible short periods of favourable growth conditions. Many micro-organisms can be "cryptobionts" - i.e. have dormant stages (spores/cysts) for survival during adverse periods such as extreme cold/hot, dessication, etc. Therefore, in the search for life elsewhere (e.g. Mars) it is probably best to seek microbial life (in the obvious absence of dinosaurs and sequoias).

What are some of the reasons for restrictive condition for life? First, is the stability of macromolecules, enzymes, membranes, etc., which are confined to limited ranges of phys./chem. conditions.

e.g. In temp ; 60-70°C - membranes (lipoproteins) melt and are destabilized.

In temp : $<0^{\circ}$C - semi-rigid/crystalline molecules results and no metabolism is possible.

Several cryoprotective measures have evolved to on one hand, survive in the extreme cold. Examples here are "antifreeze" proteins which permit supercooling. In extreme heat resistant organisms, thermostable protein and nucleic acid association exist.

In any consideration of limits to life, the availability of H_2O must be paramount. H_2O in liquid form (sat. vapor maybe) is essential for most microbes at least at some time for: solute/solvent concentration regulation; buffer (pH) control; all metabolic and growth related chemistry. Most bugs survive but don't grow well under extremes. However, some strictly adapted species have evolved (Fig. 16).

Antarctica and deserts on Earth have provided natural laboratories for extreme conditions. They have also provided an object lesson concerning our expectations for life on Mars. Antarctic interior valleys exist, which are completely arid, ice-free, and subject to very little precipitation. The diurnal mean soil temperatures ranges from +5°C to -5°C in the summer and then

Figure 16

Limits and Essentials of Microbial Life

① Water Activity

$A_w = P/P_0$

P - vapor p. of given solution
P_0 - " " " pure H_2O
at given Temp.

eg.	Blood plasma	0.99	37 °C
	Sea water	0.98	25 °C
	sat. NaCl	0.75	25 °C

② Ranges & Limits

	"Normal"	"Extremes" →	Examples
Temp. -	20-45 °C	0-100 °C	Various bacteria { psychrophili thermophilic
Press. -	1 atm +	600-1000	marine microbes
pH -	3-9	0.5-1 11-12	some bacteria some algae
salinity -	1-2 %	→ 25 %	halophilic bacteria
Aw -	0.95-0.99	0.65	" "

"Absolute" limit?

locally only. The mean annual Aw ~ 0.50, which is considered "sterile". This was considered the closest approximation to Mars' surface conditions which we could expect on Earth. At the time the space package for Mars was designed it was not known that bacteria and algae were present as "Endolithic" forms. These microbial communities survive in quartz/sandstone pockets, where light is sufficiently concentrated for photosynthetic activity and H_2O condensation. The organisms grow well and survive at ~$4^{O}C$, cohabit and recycle organic products. In fact, rapid hydration and high temperatures are lethal to some endoliths such that they did not survive earlier tests of detection.

The Search for Life on Mars (The unfinished experiment?)

To date we reviewed some of the essential basics of Earth life in terms of both, the evolution of Earth as a unique planet in the Solar System and in defining universal criteria to test for similar life elsewhere. The historic Viking 1 and 2 landings on Mars (July and August, 1976) provided the first direct opportunity to look for life on other plants. This was "exobiology" coming of age.

Some general relevant features of Mars in this connection are:

1. The planet is of the "terrestrial" type, but only about ½ the size of Earth.

2. Mars experiences days (~ 24½ h long) and seasons in a year, about 2 times as long as Earth's. Mars has a tenous atmosphere (1/1000 Earth's) composed of mainly CO_2. Mars has a heterogeneous surface and distinct polar, temperate and equatorial regions. Until the actual Viking landing, the best questimates of surface conditions were approximately similar to those of the Antarctic dry valleys. The temperature conditions are generally below $0^{O}C$ most of the year, with locally warmer periods (~$+10^{O}C$). Water would be mostly

present as permafrost, and the effective Aw = below "life" levels most of the time, with possible locally higher levels in the summer. The atmosphere was thought mainly CO_2 (traces of other gases) and pressure of 8-10 millibars (lower than Mt. Everest or about 1/1000 sea level). The Viking life detection experiments were designed with above scenario in mind, plus summary of basic aspects of Earth micro-organisms:

(a) all in essence assimilate CO_2 for primary biosynthesis.

(b) all require H_2O for active metabolism.

(c) the primary energy source is light (photo-energy).

(d) all metabolize above freezing point of H_2O.

(e) all produce CO_2/O_2/? as metabolic by/end products.

(f) all "heat" sensitive → i.e. killed much above ∿100°C.

The Viking Bio-package was broadly designed to assay for bio - activity under expected "optimal" life conditions, i.e. if Mars bugs are dormant (e.g. spores, seeds, etc.) then by changing conditions to equivalent of "spring", i.e. raise the temperature, add H_2O and nutrients and mix. Thus, by providing Mars bugs with a ready-made "Chicken Soup", perhaps their presence would be detected in a manner giv~~e~~ing clear cut yes/no answers to whether or not life is there.

The Viking bio package was equipped to receive soil samples for 3 types of experiments:

1. Carbon Assimilation - seeking evidence for photosynthesis or some other equivalent primary energy assimilation mechanism.
2. Gas Exchange
3. Labelled release

} seeking evidence of metabolic activity, mainly Respiration.

The landers were deployed at 2 sites on Mars: Viking 1 at 22°N lat.:- "equatorial" region; and Viking 2 at 47°N lat.:- "temperate" region. Both sites were superficially similar, with rock, sand, dry environments. The ambient temperatures ranged from -80° to -30°C (locally higher) and the ambient atmospheric composition was: CO_2 (95%), N_2, Argon, traces CO, O_2. The rationale and results of the Carbon Assimilation experiment are shown in Figs. 17 & 18. The initial conclusions about the Carbon Assimilation experiments were as follows:

1. Weak positive indication of bio-activity was possible.
2. The observed activity was heat labile (i.e. sterilizable), and H_2O sensitive, maybe light stimulated.

<u>The Gas Exchange Experiment</u> had a similar basis and rationale: feed the "microbes" a liquid nutrient medium containing amino acids, sugars under "optimal" growth temp. 10°C, and look for evolution (release) of CO_2, O_2, N_2, etc. as metabolic by-products, i.e. respiration (Fig. 19a). The conclusions of this experiment were that the results are probably not indicative of bio-activity. The kinetics of CO_2 evolution were not bio-like and probably due to oxidation of organics in medium by chemicals in the soil, e.g. peroxides + Fe oxides + nutrients + H_2O. Simulated soil experiments on Earth gave similar results (Fig. 19b).

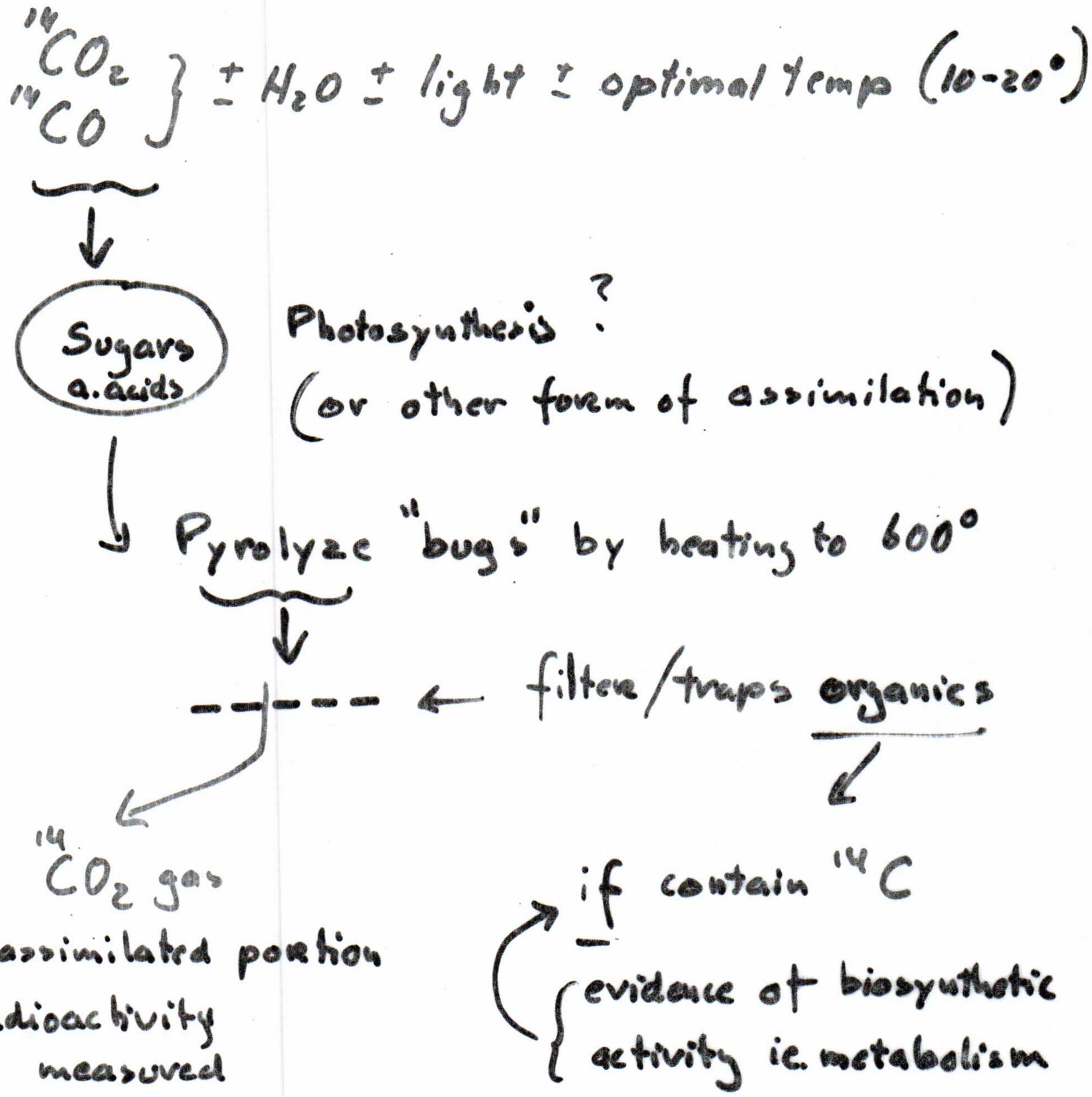
<u>The Labelled Release Experiment</u>. This was in many ways the most clear cut (in principle) experiment, i.e. with control Earth samples providing no problems and unambiguous yes/no. The experiment is the most "biological" in that active cycle (live bugs) and sterile cycles (i.e. killed bugs) could be compared for long term/short term trials and under different conditions of temp., pH, humidity, etc.

The rationale here was as follows: feed the bugs ^{14}C-labelled nutrient medium and measure the release of $^{14}CO_2$ as evidence of respiration. Rates of CO_2 release was monitored and compared to Earth soils. As controls heat sterilized soil samples were examined since no known inorganic reactions can

Fig. 11

Rationale for Viking Carbon uptake Expmnt.

① Supply Martian "microbes" with:

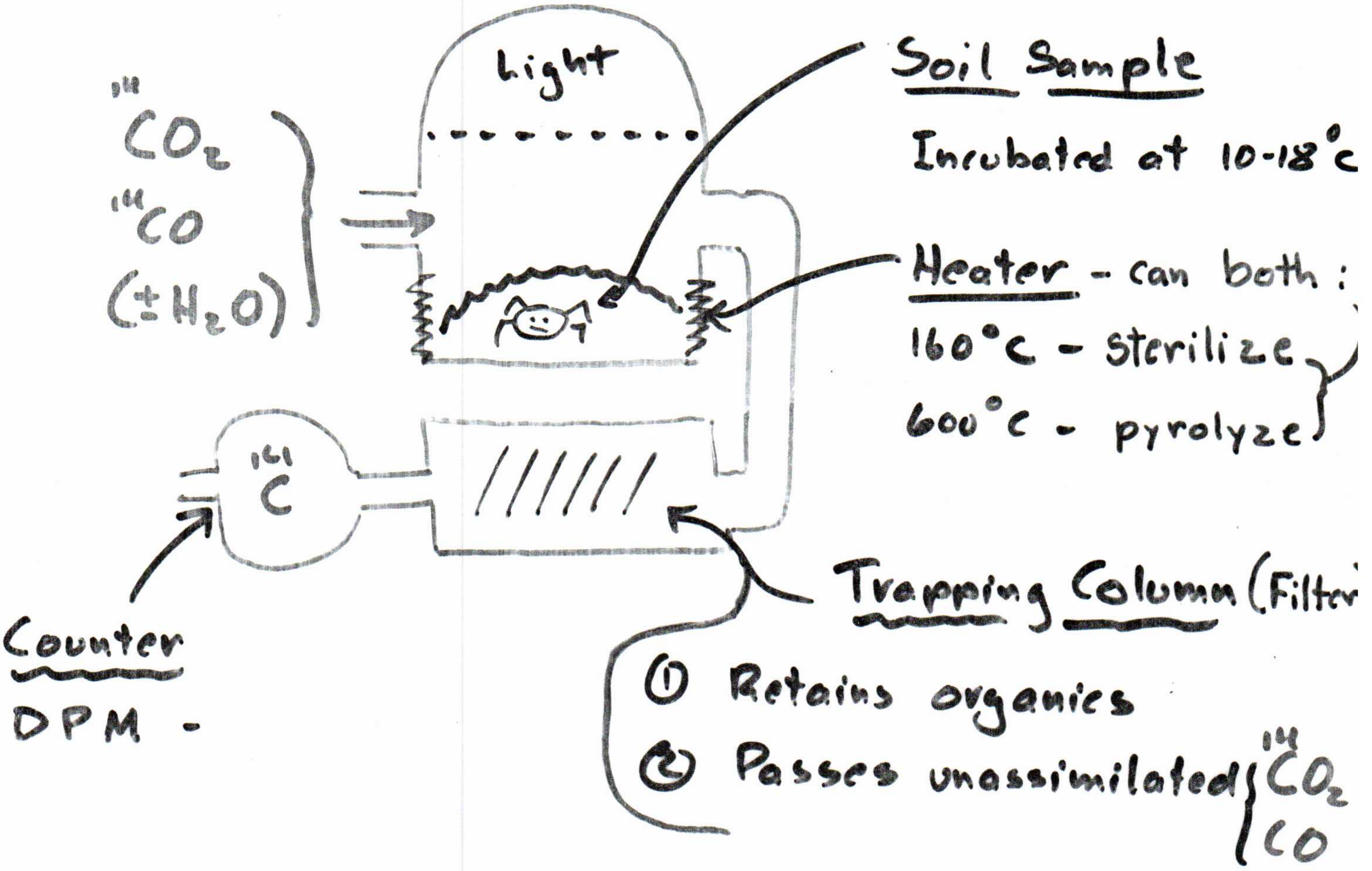


② Repeat above but sterilize soil first to kill "bugs"

③ Repeat longer/shorter times for assimilation, etc.

Fig. 18

Viking Experiment - Carbon Assimilation (Pyrolytic Release



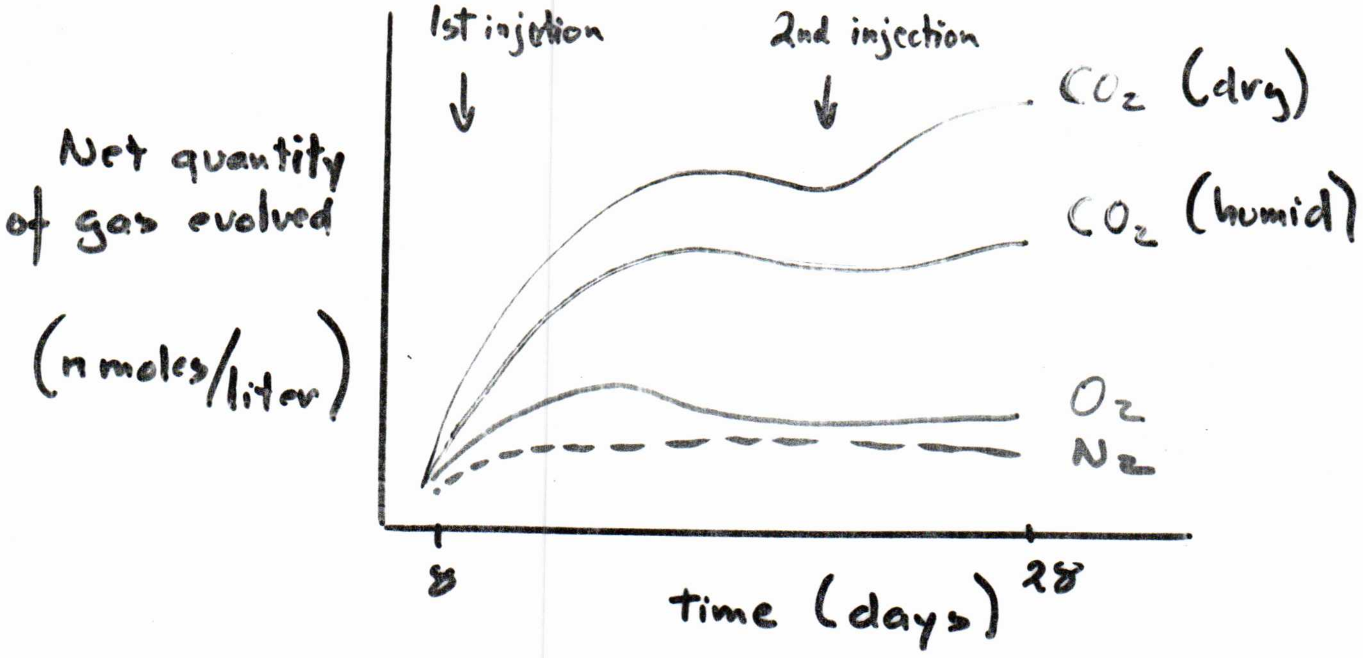
Results

Conditions of assay	DPM counted	
	$^{14}CO_2$, CO	"Organics"?
Active cycle (light, dry)	7,400	96 ± 1
Control " (sterilized soil)	7,650	15 ± 1
Active cycle (light, wet)	12,500	3 ± 1
Active cycle (dark, dry)	13,000	7 ± 1

(counts after correction for background)

Viking Experiment - Gas Exchange

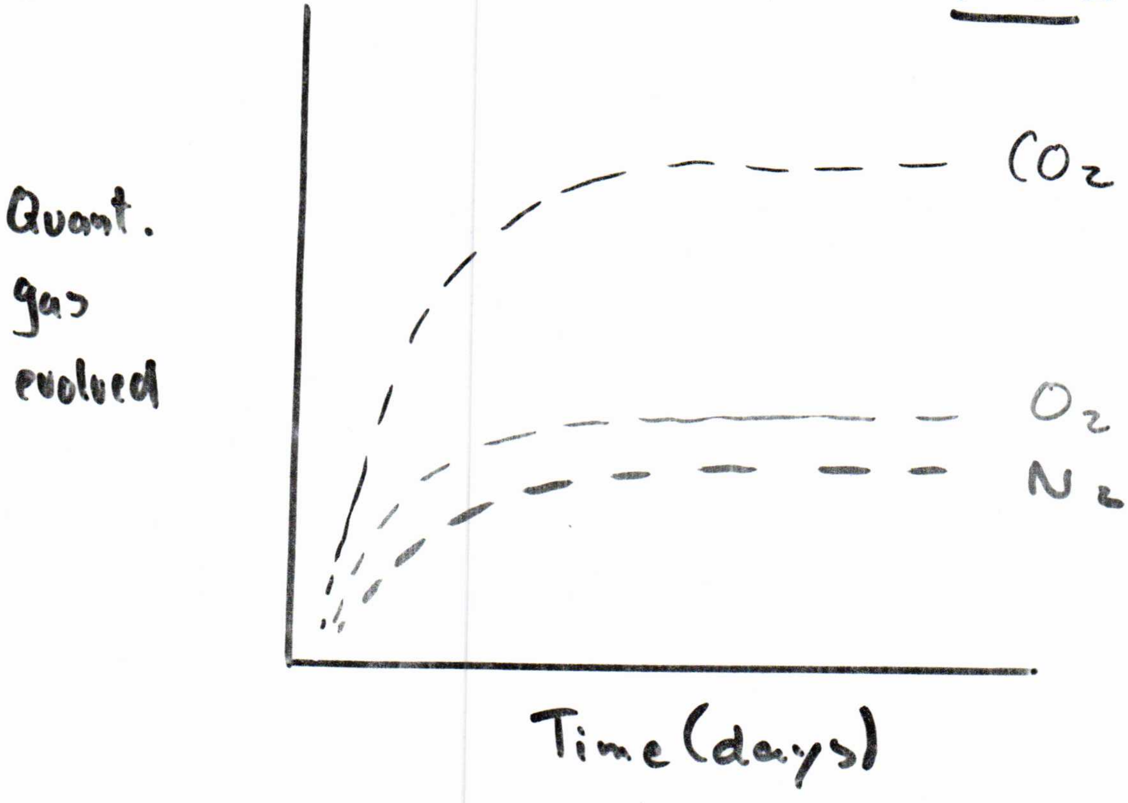
(Fig. 19 a)



Simulated Martian Soil - (Amboy + Ca superoxide)

(Fig. 19b)

All in dry mode



be "killed") (Figs. 20, 21).

The conclusions of this important experiment were:

1. a weaker but very similar activity to Earth soil was indicated, although rates of CO_2 evolution were very much slower.
2. the activity is inactivated by 160°C, i.e. killable, unlike any known chemical (inorganic) activity on Earth.
3. a weak, cyclical, residual activity was retained at elevated temp. 50°C, i.e. suggesting partly killed bugs?
4. all the above are not readily explained in any non-biological ways.

Is there life on Mars then? Probably not but, this is clearly an unfinished experiment. However, a non-biological explanation is probable for most of the Viking experimental results because:

(a) the soil is unduly "oxidizing" due to the presence of Superoxides H_2O_2, Iron oxides Fe_2O_3, etc..

(b) the high UV flux on Mars, probably decomposes any possible surface organisms and organic compounds (the precise intensity of UV flux not clear yet)

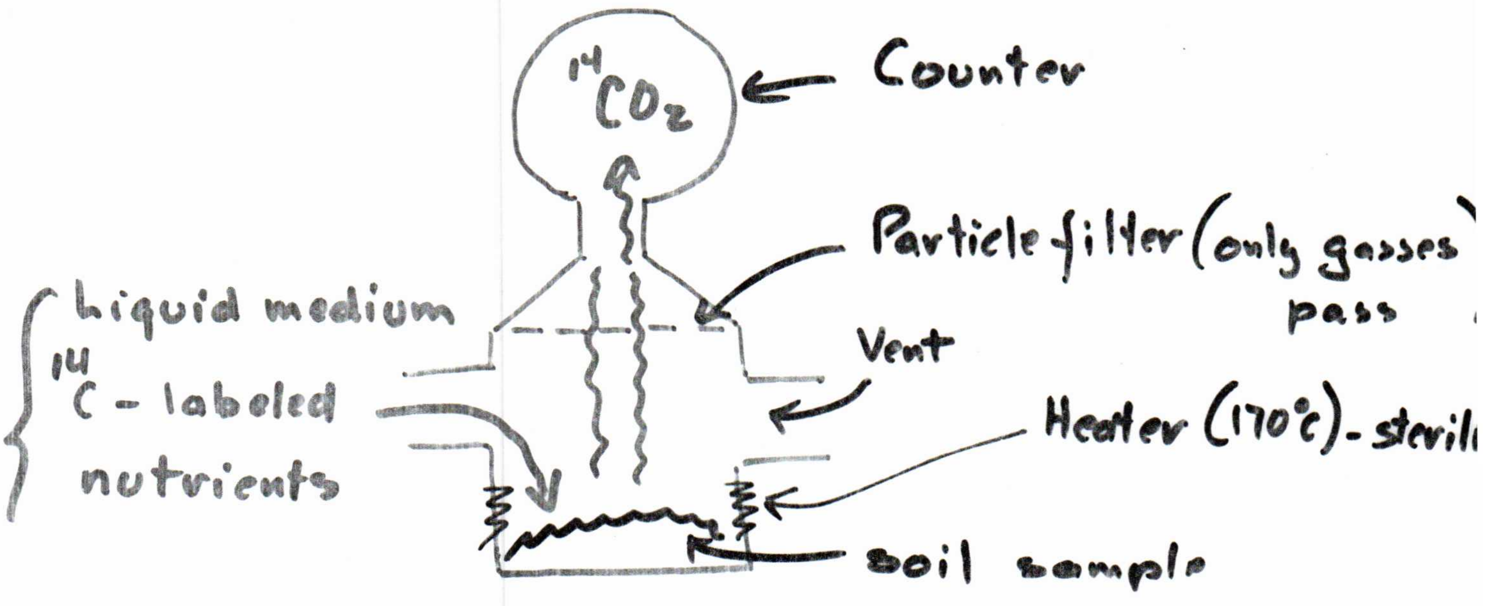
(c) most simulation experiments carried out on Earth using Viking-type assays indicate that most gas evolution is likely inorganic.

(d) independent analysis (mass spectr., gas chromat.) indicate that only very minimal numbers of organisms could possibly be present in the Martian soil.

In retrospect of course, and with the benefit of new data, several questions and possibilities remain unclear. If the Mars surface chemistry is radically different from Earth's and if life did evolve there in the past, then presumptive bugs must have adapted to the very harsh environment such that:

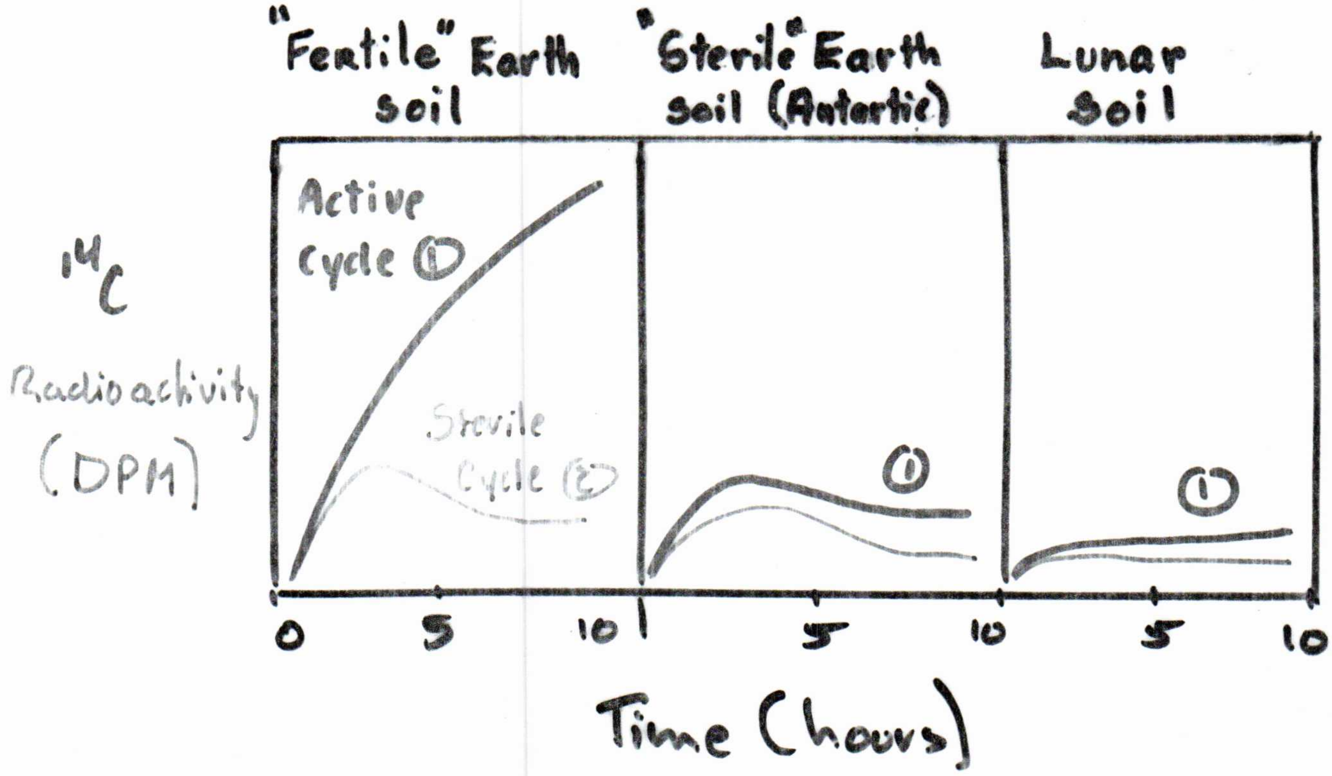
1. bugs are present but only in minute and scattered pockets in closed

Viking Experiment - Labeled Release (Fig. 20)



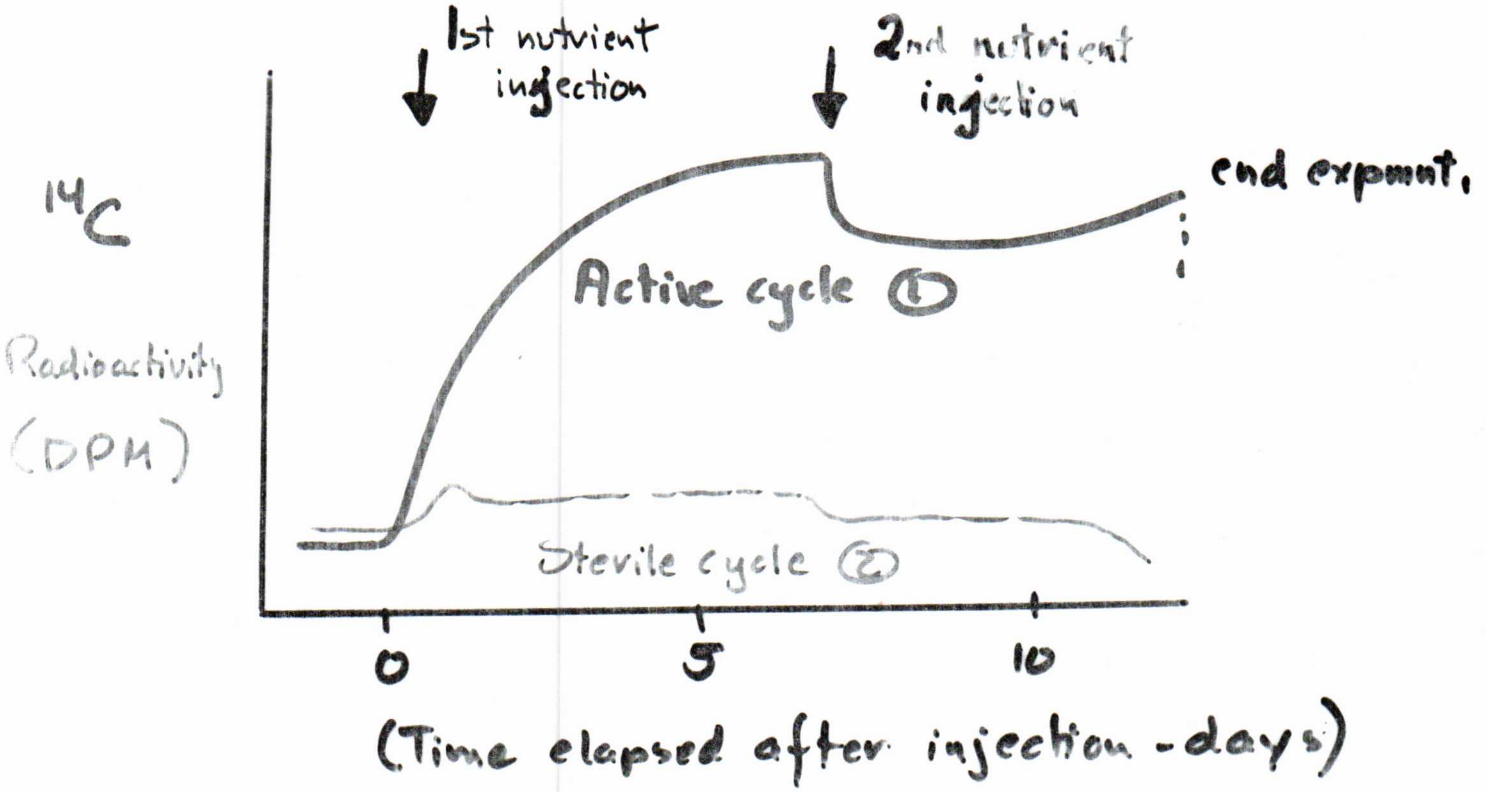
All sample split for: ① Active cycle
② Sterile cycle

Control Results with non-Mars soils

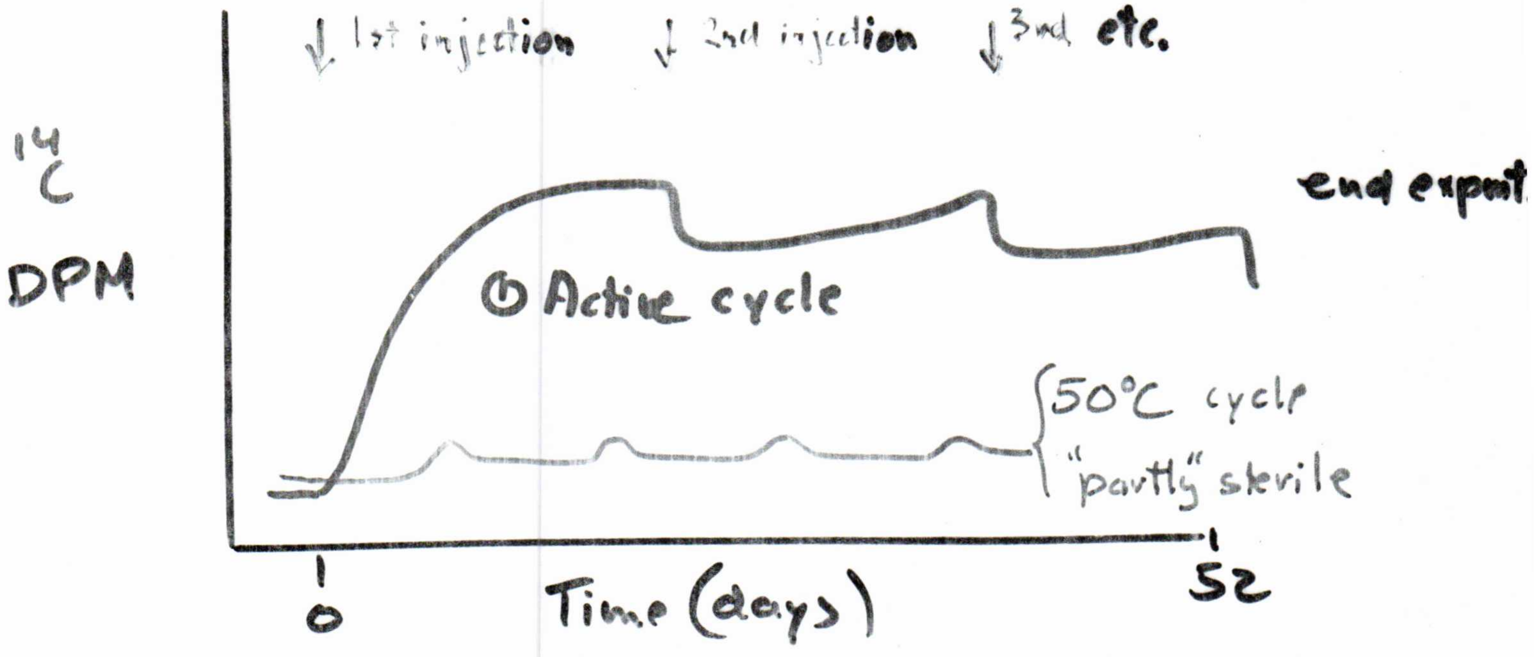


Viking Experiment - Labeled Release Results (Fig. a

Mars soils → similar from all sites tested



Extended Experiment



system needing only occasional energy input (temp, light) from exterior.

2. the Martians metabolize best and grow at temperatures and Aw much lower than thought possible.

3. excess H_2O maybe detrimental.

For future exploration of Mars then, if the planet is akin to the Antarctic (only much more extreme - colder, drier, UV) perhaps we should explore areas near the polar caps - where local pockets of H_2O vapor may be available. Most interesting, however, perhaps we should look for evidence of fossil life or "cryptobiological" forms. It is possible that past/future conditions are quite different and that at times in the past, Mars may have enjoyed more moderate surface conditions. For example, calculations indicate that if the polar caps were evaporated completely enough frozen H_2O and CO_2 would be released to make the Martian atmosphere as dense as the Earth's today. Under these circumstances liquid H_2O would be present on Mars. Carl Sagan suggests that Mars is in an ice age now and that possibly due to orbital changes and axial tilt (precession) etc., a ∿50,000 yr cycle exists on Mars whereby temperatures + greenhouse effects may be raised sufficiently to provide life conducive conditions every 25,000 years. Conceivably, indigenous life could have evolved to adapt to such cycles. The best bet for now then, is if life evolved when Mars conditions were most favourable in past, some organisms may have evolved and adapted to present extremes and only pockets or fossils of a few microbes remain.

Other Life Systems and Alternate Biochemistries

Any examination of questions related to the origin of life and its potential abundance in the Universe must ask whether the biochemical bases of terrestrial life are unique or whether alternates are possible. For instance, can life evolve only along a carbon-water basis or can other elements/compounds do just as well, e.g. Silicon? Is life, even totally alien life, restricted to planetary bodies with fairly defined and narrow temperature/energy regimes, or can other systems or environments serve as platforms for life? Let us examine some of these aspects in more detail. Why carbon based life? Because:

1. Carbon has a valency of 4 and bonds with itself to form an infinite variety of different molecular shapes and forms.
2. Carbon combines with a larger number of elements than any other (H, O, N, P, S, etc.) to form compounds in solid, liquid, and gaseous states over a fairly narrow range of temperature and pressures.
3. In short, no other element is as <u>versatile</u> as carbon. Silicon, which also has a valency of 4 tends to combine with other elements (notably O_2) to form large, but rigid polymers of restricted variability and versatility.

Why H_2O as a basis for life? Water has been called a universal solvent, (i.e.) a solvent for more types of molecules at broad temp/press ranges than most other compounds. Water has the unique property of forming ice on top, ∴ protective of the liquid medium underneath. This is clearly important for survival of any living organism in a liquid medium. However, alternates to water exist. For example, ammonia can act as solvent similar in some ways to water ($H_2O = H^+ + OH^-$, $NH_3 = H^+ + (NH_2)^-$). NH_3 is also similar to water in terms of such things as molecular polarity, acid/base properties, etc. Ordinary proteins have been found stable in liquid NH_3, which however, is liquid at ∿-30°C and/or high pressure. Therefore, the thermodynamics of ionic (Redox) reactions in liquid NH_3 would be relatively sluggish or slower compared to H_2O. Although, theoretically at least, ammonia is not excluded as a solvent for life, high (even low conc.) of NH_3 is toxic to all Earth organisms (0.01M upper limit for most tolerant microbes). Thus any ammonia life would have to be different in that respect at least, although O_2 could similarly be toxic to alien life.

Surprises are still possible, however, even on Earth. The best example here is a recently discovered group of primitive bacteria called Methanogens. These are thought to be among the most primitive groups of microbes known. They are strict anaerobes (any O_2 is lethal), slow growing as group symbionts (like Antarctica bugs). E: assimilation is varied:

$$\begin{tikzcd}
 & ATP \\
2\,H_2R + CO_2 \arrow[r] \arrow[ru] & CH_4 + H_2O \arrow[d] \\
C^-/H^+ \text{ donor} & \text{can be } H_2 \arrow[l]
\end{tikzcd}$$

Thus they must live under very reducing conditions and probably would be happy in a completely O_2-free atmosphere, i.e. CO_2, H_2O, CH_4 like the primitive Earth?

Life in the Solar System

Excluding the Earth and Mars, are there any other potential abodes of life or even pre-life conditions in the solar system? Even at a casual glance, the planets Mercury and Pluto, as well as most other minor planets and satellites of other planets, are clearly out of bounds to life. Any of these bodies have surface conditions which are either too hot or cold, too dry and airless, too close ~~for~~ to the sun or too far, etc. to permit life to have ever evolved there, now or in the past.

Venus is of some interest, however, because in many ways it is Earth-like with respect to distance from the sun, size, etc. The atmosphere and surface conditions on Venus are such, however, to probably exclude life completely. The atmosphere is mainly CO_2 (97%) with traces of N_2, H_2O, NH_3 and H_2SO_4 clouds. The atmosphere is very dense (90X pressure of 1 Earth atmosphere on surface), cloud covered and probably represents a runaway "greenhouse" effect, which partly explains the excessively high surface temperature $>500^{\circ}C$ observed. It has been suggested that possibly non-biological synthesis of some organic compounds might take place in the upper levels of the atmosphere of Venus, where temperatures around 273°C may prevail. Life as we know it, however, is not present on Venus.

The Jovian (Jupiter-like) Planets

Jupiter, Saturn, Uranus and Neptune may be of considerable interest in terms of possible "prebiotic" conditions prevailing at least at some level in the atmosphere. These are non-terrestrial planets, i.e. there is no real surface but a presumed transition from: gas/liquid/solid. Various "models" of Jupiter in particular exist. This is the best studied of the giant planets and most models agree that:

1. Very "reducing" conditions prevail, i.e. H_2, He 80-90%, traces of CH_4, NH_3, H_2O, H_2S are present.
2. Temperatures in the "biotic" range are found at some level.
3. Abundant energy is available, including UV, electric discharge and thermal.
4. Aw, pH, etc. may be regionally within Earth levels.
5. Extensive convection and recycling of atmosphere takes place (Fig. 22).

Overall this general scenario suggests that portions of Jupiter maybe a giant spark-discharge chamber where organic synthesis can take place. Many simulation experiments have been done using Jovian atmospheres in a Miller apparatus to synthesize various complex organics (Fig. 23).

The foregoing has raised speculations about a Jupiter "Life Zone" or "biotorus" in the upper atmosphere, where recycling and convection could support droplets of microbial organisms (Fig. 24). Clearly this is very speculative and unlikely, but interesting nontheless for future space craft explorations.

Fig 22

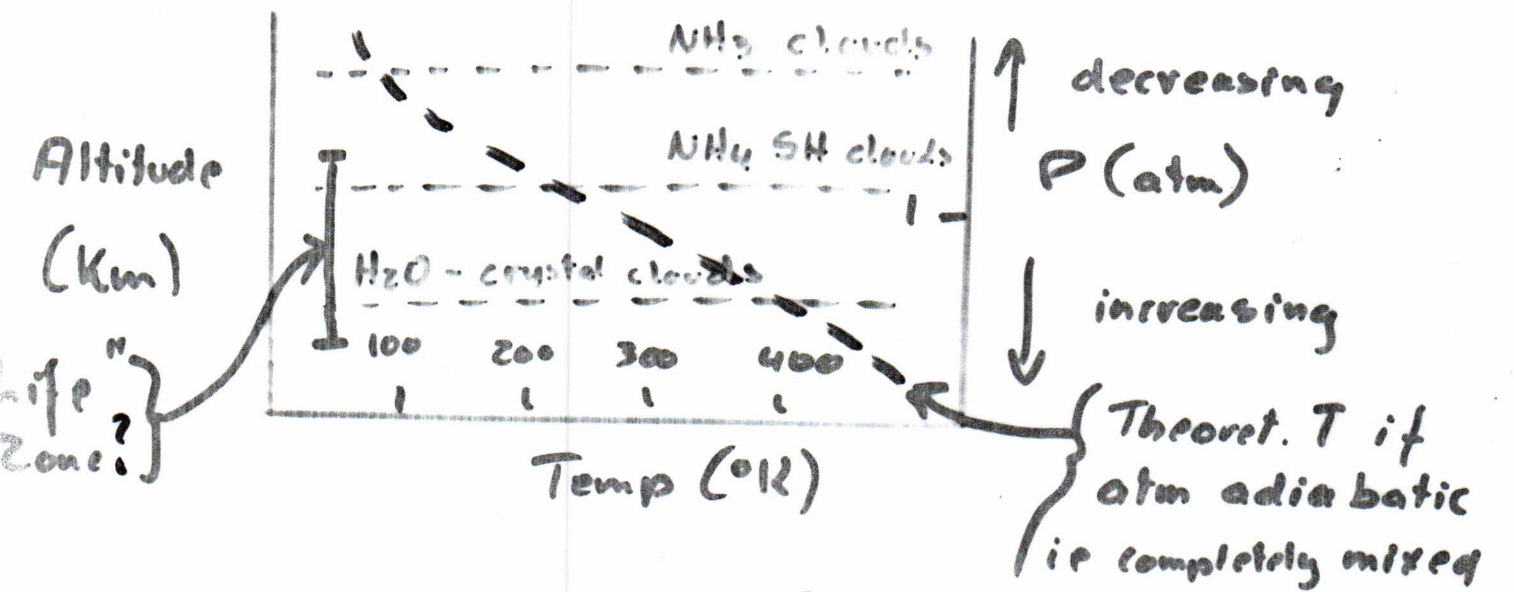
Atmospheric Composition of Jovian Planets

(T. Owen

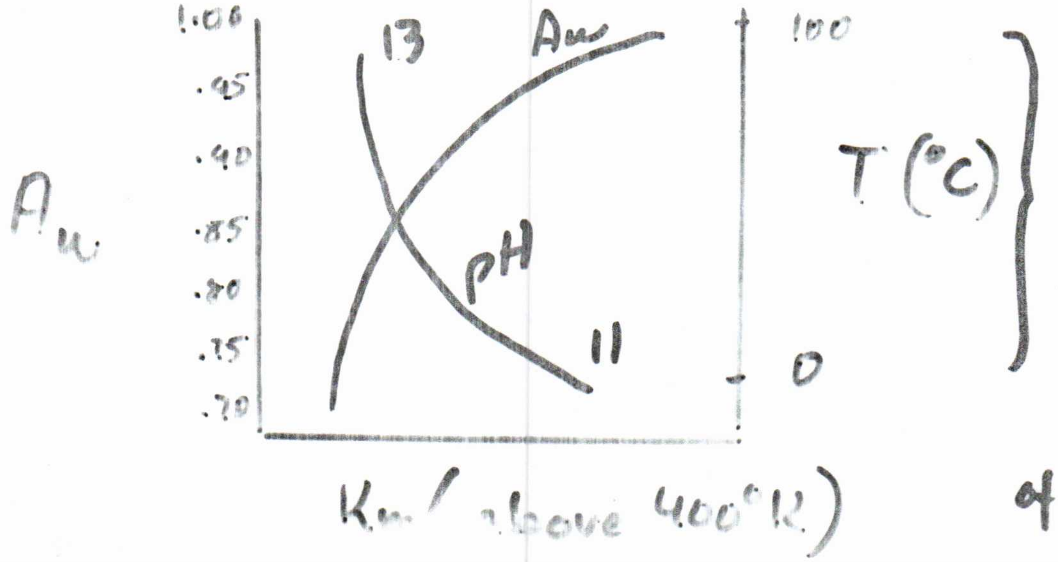
① Decreasing abundance of gases:
(Some measured, some estimated only)

H_2 } 80-90 % — C_nH_n } traces only
He — HCN
CH_4 — H_2S } {Very "Reducing" atm
NH_3 — H_2O ← {main source of O_2

2) Models of Upper Atm. Structure (Jupiter)



3) Theoret. Water Activity (Horowitz)



} "Life" range?

Aw = OK (>0.62)

pH = high
but in range
of many micro-
organisms

[Figure 23]

Organic synthesis from Jovian atmosphere simulations in laboratory experiments

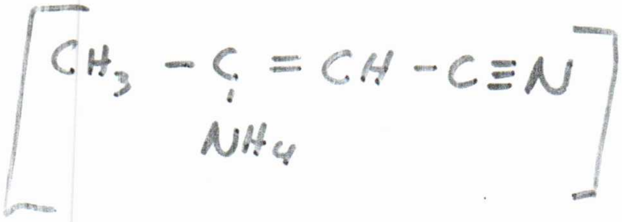
Starting mixes (various)

$$\left.\begin{matrix}CH_4\\C_2H_6\\NH_3\\H_2S\\H_2O\end{matrix}\right\} \xrightarrow[\text{spark discharge}]{\text{U.V.}} \left\{\begin{matrix}95\%\ S\\ \\ 5\%\\ \text{complex organic solids}\end{matrix}\right\}$$

Red
Brown
Orange
colours

tars, nitriles etc.
(polymers)

hydrolysis



$$\left[CH_3-\underset{NH_4}{C}=CH-C\equiv N\right]$$

Various nitriles

① Pre-cursors of Amino acids in most abiological synthesis reactions à la Miller

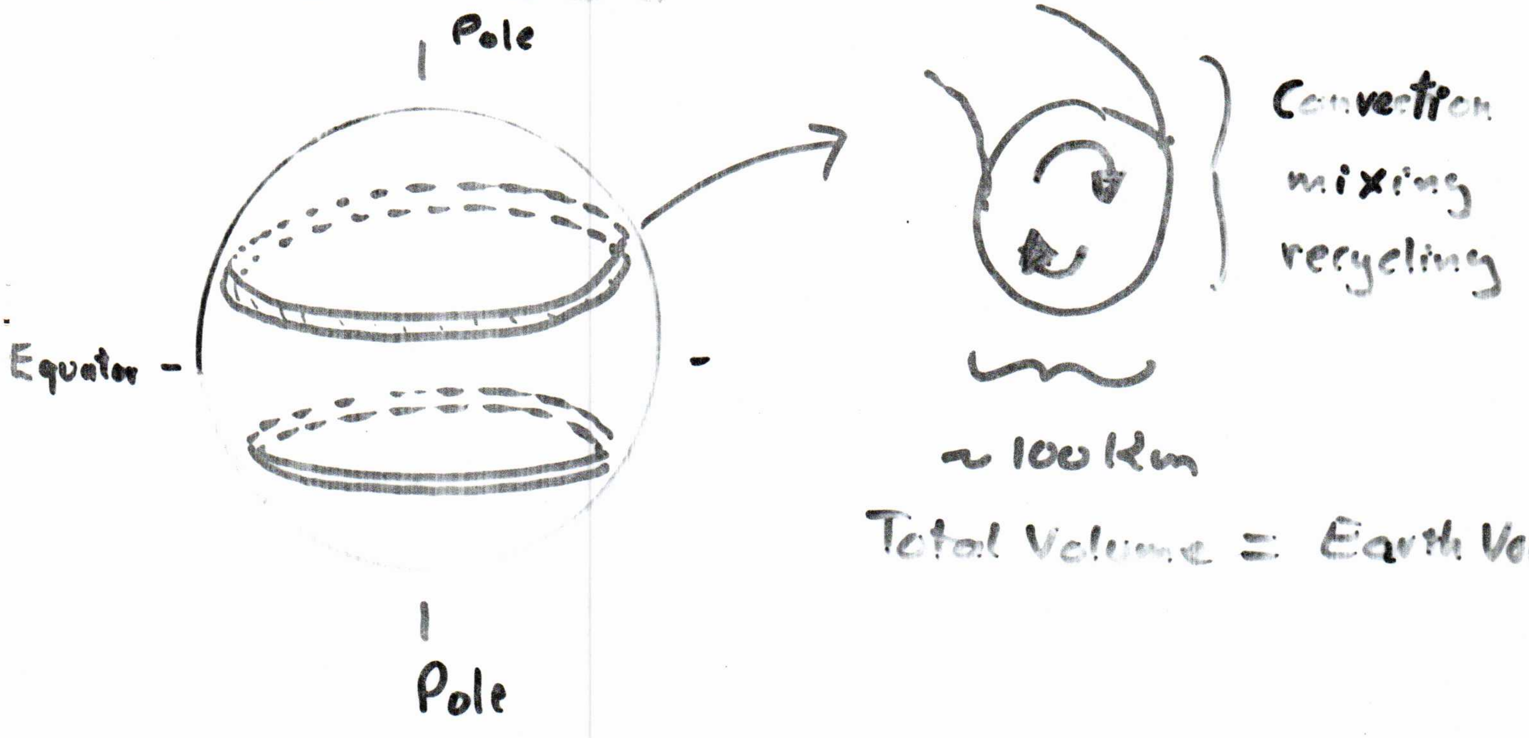
② Pyrolytic products of amino acids peptides nucleosides etc.

"Life" on Jupiter? Fig. 24 (MacElroy)

① Temp – Pressure Relationships in Jup. Atm.

Distance from planet's center (Km.)	Temp. (°K)	Press. (atm.)	Chemical Comp.
70,600 – 70,560	?		He H_2
70,500 ~	100 150	0.001	NH_3 ice NH_4SH
70,400	~ 285	0.2	H_2O
70,300	~ 400	0.5	NH_3, H_2O, CH_4 C_2H_6

② The "Biosphere"? Bio-Torus



Total Volume = Earth Volu

Titan

This is the major satellite of Saturn. It is also the largest satellite in the solar system, being about 3/4 the size of Mars. It is small and also very distant from the sun. However, it is a solid, terrestrial-type body with a distinct and extensive atmosphere. The atmosphere is a yellow-red colour (press $\cong$ 0.4 Earth) and CH_4 (methane) has been detected for sure - maybe also H_2, He, NH_3, H_2O. Several models for Titan's atmosphere have been put forth, ranging from a completely frozen-out atmosphere to liquid NH_3 on the surface. Temperatures in the 150-200°K (maybe higher) might exist and therefore this is clearly of prime interest for future space craft exploration, because:

(a) maybe "prebiotic" chemistry prevails on an accessible planetary body;

(b) conditions are probably too harsh for life but a natural laboratory may exist here to test many alternate biochemistries.

If liquid ammonia is present then a great testing ground exists.

Alternate Origins?

It is generally accepted that life on Earth evolved as outlined previously in the following sequence:

1. 4.5-4 BY ago was the Earth's condensation and cooling period;
2. $\sim$4 BY(?) prebiotic synthesis began and was followed by chemical evolution leading to protocells then true cells; about 3.5-3.7 BY ago, the first Prokaryotes; and 1 BY ago to the first Eukaryotes.

If this time scale is correct, life evolved from molecules to cell relatively "quickly", i.e. if it took 2.5 BY to evolve Eukaryotes from first the bacteria, how come it only took $\lesssim$ 1 BY to evolve life in the first place? Furthermore, how come all existing life forms have fundamentally the same biochemistry, architecture, info transfer, etc? This implies that the first cells reached a very complex level quickly and why are no alternate/parallel systems evident at least part way? Why, if nature likes to experiment, did she not do more of it here?

Obviously, there is no clear-cut answer to this - but some alternatives exist, including:

1. Panspermia - this concept holds that life in the form of spores (from exploded planets?) drift through space and 'infect' a sterile early Earth. Panspermia could be random, directed/accident by aliens (space probes), God. Clearly this is not a satisfactory answer to the origin of life since it poses as many questions as are answered.
2. "Head start" theories - these have gained more credence in light of the advent of "Astrochemistry". Many complex organic molecules have been detected in interstellar clouds, including glycine, the simplest amino acid. This indicates that the abiotic synthesis of complex organics is possible and that these are not destroyed even in outer reaches of space. This might provide a reservoir of building blocks for life, both locally and throughout the universe.

Is there any evidence for this idea? The best evidence is from a class of meteorites called "Carbonaceous chondrites". These are rare meteorites, ∿4.5+ BY old and are considered remnants of earliest stages of solar system formation. They are rich in Mg/Fe silicates, sulfur, and ∿5% organics. This provides good evidence for differentiation of solids in early solar systems and for the prebiotic synthesis of organic compounds. The Murchison (1969) meteor in Australia contains most of the amino acids of biological significance and others as well.

Together this type of information has been integrated by several authors into the general scheme outlined in Fig.25. Whether this "head start" idea for life on Earth (and by implication of other planets in the universe) hold true of course, awaits further exploration of other elements in the solar system.

Figure

(Hoyle and Wickramasinghe theory)
and others