Origin of life and energy

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GLOSSARY

dehydration condensates: polymers in which the monomer linkages are equivalent to the removal of a molecule of water between the joined monomers.

Gibbs free energy: a type of thermodynamic energy containing internal energy, entropic energy and pressure volume energy that is applicable for systems at constant temperature and pressure.

macromolecular: pertaining to molecules of large size having masses in the thousands to millions of daltons (one dalton is one atomic mass unit). All biologically important macromolecules are polymers, molecules made from linking together monomers by means of dehydration linkages. There are three major types: proteins made from amino acid monomers, polysaccharides made from simple sugars and polynucleotides made from mononucleotides.

micells: self-assembled monolayered closed structures made from fatty acids dissolved in water.

phosphate: a molecule made from phosphorus, oxygen and hydrogen that is an ion when dissolved in water with the formula HPO_4^{2-}.

protein biosynthesis: the complex mechanism by which proteins are synthesized, amino acid by amino acid, in accord with a sequence of nucleic acid bases in a messenger RNA molecule that has been transcribed from a DNA gene. Many proteins and RNA molecules are involved in the apparatus that performs this function.

redox energy: the energy of electrons transferred with them during electron transfer reactions.

stromatolites: layered fossil rocks believed to be the remains of mats of bacterial colonies topped and fed by phototrophic organisms, perhaps the ancestors of contemporary cyanobacteria.

thioester: an energy rich compound involving sulfur covalently linked to the carbon of a carbonyl group (see section VI).

vesicle: a bilayer, self-assembled, closed structure made from glycerophospholipids possessing two fatty acid side chains.

I. Definition of origin of life and energy

The phrase **origin of life** refers to those natural geophysical processes that may have occurred on the primitive Earth some 3.5-4.0 billion years ago that gave rise to life. Presumably there are no extant representatives of the earliest forms of life since they were surely driven to extinction by more advanced forms very early in the evolution of life. Fossils do not preserve *macromolecular* structures inside cells so that no direct fossil evidence exists. Even putative cellular fossils from 3.5 billion years ago, the *stromatolites*, are not absolutely proven to be what they appear to be. On the other hand, their similarity to contemporary formations deposited by bacterial mats topped with cyanobacteria, that may not be too different from

the primitive organisms of 3.5 billion years ago, suggests that the metabolic and *protein biosynthesis* macromolecular machinery of contemporary cells was already developed that long ago. How these macromolecular structures came into being is the problem of the origin of life.

In this context **energy** refers to a variety of forms, both chemical and physical that are relevant in the context of the origin of life. These include: 1) fusion energy released during the formation of the nuclei of elements during stellar nucleosynthesis; 2) ultraviolet (UV) light energy of the sun that excites electrons into excited states thereby providing them with *redox energy*, for example during the oxidation of iron from ferrous iron (Fe^{2+}) to ferric iron (Fe^{3+}); 3) *thioester* compounds that are rich in *Gibbs free energy* so that they spontaneously engage in a variety of reactions and syntheses; 4) *phosphate* compounds that serve to activate intermediates in the synthesis of polymers; and 5) thermal energy or heat. The problem for the origin of life and energy is to understand how these different forms of energy are connected and transduced from one kind to another and promote the origin and evolution of the macromolecular processes of living matter. In contemporary organism there is also the fundamental importance of chemiosmotic proton energy, but this form of energy likely evolved relatively late in the origin of the first cells.

II. Stellar nucleosynthesis of the elements

Understanding why life is made out of the elements that are observed to be most abundantly present in organisms is aided by considering the origin of the elements during stellar nucleosynthesis. These elements are H(ydrogen), C(arbon), N(itrogen), O(xygen), P(hosphorous), S(ulfur), Cl[chlorine], Na[sodium, but from Na(trium)], Mg[magnesium], K[potassium, but from K(alium)], Ca(lcium) and Fe[iron, but from Fe(rrum)]. While several other elements are essential for many contemporary organisms, such as I(odine), Se(lenium) and Co(balt), primitive life almost certainly could have arisen based upon the primordial dozen elements above. These elements are the available building materials for life because they include the most abundant naturally occurring elements.

During the lifetime of a typical star, tremendous changes in pressure, mass density and temperature take place inside the star. Temperatures between 10^7 and 10^9 K occur, as well as densities from 10^2 gm/cm^3 to 10^5 gm/cm^3 (ordinary liquid water has a density of 1.0 gm/cm^3). These extreme conditions promote a series of nuclear fusion processes that are organized around the nucleus of helium, called an alpha particle, that contains two protons and two neutrons. This is a particularly stable configuration of nucleons (protons or neutrons) and its formation from free, but very hot

protons (the H nucleus) releases vast amounts of energy, thereby driving the formation process. Subsequent fusion involves the incorporation of more protons into the alpha particles and tends to give rise to nuclei that are simple integer multiples of alphas, the nuclei of C, O, Mg, Si(licon), S, Ca and Fe. Notice the presence of Si and the absence of N, P, Na, K and Cl. The processes of synthesis of the alpha multiple nuclei generate these additional elements, albeit in lower amounts than the true alpha multiples. As will be seen in section VIII, the d-orbital properties of Si make it unsuitable for the basic metabolic constituents of cells, and it instead has a very strong tendency to combine with O to form enormous three dimensional inert silicate compounds such as quartz. Fusion energy release leads to the elements of the first four periods of the periodic table, the most abundant elements in the cosmos, and these are the building blocks of living matter.

III. Relative abundance of the elements

By weight, ¾ of the Earth's crust is made of O (50%) and Si (25%), primarily as silicates. The oceans are mostly O (86%) and H (11%), primarily as water. While abundance is important, fitness is just as important. Si is 135 times more abundant than C anywhere on Earth even remotely suitable for life, and shares with C the tendency to form four covalent bonds, but it is highly unfit as a constituent of living matter

compared to C. C can combine with other types of atoms to form myriads of small molecules, some of which involve double bonds. It combines to make strong stable bonds. Si however combines most strongly with O to make highly polymeric inert structures called silicates. Si-Si bonds are highly susceptible to attack by H_2O, NH_3^+, and O_2 unlike C-C bonds which are stable. Only when structural material in the form of spines and spicules is used by living organisms such as cacti and sponges, does Si actually occur in living matter. The metabolic pathways, however, are devoid of Si, using C compounds instead. These differences between C and Si have to do with where they are in the second and third periods of the periodic table respectively. More will be said about this in section VIII.

H, C, N and O make up about 99% of the constituents of all living organisms. As members of the first and second periods, they are the most abundant elements and they combine to make short, strong, stable bonds. P and S, while less abundant are nevertheless more abundant then still heavier elements and because they reside in the third period they possess unfilled d-orbitals. These orbitals confer on them the ability to engage in transfer reactions that mediate energy changes. S, in the form of the thiol group, –SH, contributes to thioester based energy transactions, and P combines with O to form phosphate that contributes to energy activations for the promotion

of polymerizations. The most stable nucleus of all, and the end product of the fusion processes in stellar nucleosynthesis, belongs to Fe. This element is ideally suited as a conveyer of redox energy. Generation of redox energy, the energy of excited electrons, is caused by sunlight.

IV. Gibbs Free energies of formation

Once the elements have formed and planetary systems have accreted around Sun-like stars, temperatures become low enough for the spontaneous formation of many types of small molecules. This spontaneity is measured by the Gibbs free energy of formation from the elements. In a spontaneous process at constant temperature and pressure the Gibbs free energy must decrease. This says nothing about rates. Without catalysts, a long time may be required before the minimum in free energy is achieved. The statement above is the Second Law of Thermodynamics expressed in terms of the Gibbs free energy rather than the entropy. This difference is caused by the difference between applying the Second Law to an system in contact with both temperature and pressure reservoirs and an isolated system, respectively.

In table I, the Gibbs free energy of formation from the elements for a variety of compounds is listed for the standard conditions of a temperature of 298 K and atmospheric pressure. The table lists the name of the

substance, its empirical formula, its molecular weight, its free energy of formation as $-\Delta G^0_{298}$ wherein the minus sign is explicitly included (a positive listing in the table means a negative free energy of formation), and its free energy of formation per gram as $\frac{-\Delta G^0_{298}}{gm}$. By convention, the elements are given the value 0.0.

Table I

Name	Empirical formula	Molecular weight (gm/mol)	$-\Delta G^0_{298}$ $(kcal/mol)$	$\frac{-\Delta G^0_{298}}{g}$ $(kcal/gm)$
acetaldehyde	C_2H_4O	44	33.4	0.759
acetic acid	$C_2H_4O_2$	60	94.7	1.578
acetate (aq)	$C_2H_3O_2^-$	59	89.0	1.508
acetly CoA	$C_{23}H_{39}O_{18}N_7P_3S$	825	89.4	0.108
adenine (aq)	$C_5H_5N_5$	135	-74.87	-0.554
adenosine (aq)	$C_{10}H_{13}N_5O_4$	267	46.46	0.174
alanine	$C_3H_7O_2N$	89	88.7	0.997
arginine	$C_6H_{15}O_2N_4$	175	126.7	0.724
asparagine	$C_4H_8O_3N_2$	132	125.8	0.953
aspartate	$C_4H_6O_4N^-$	132	167.4	1.268
ammonium ion (aq)	NH_4^+	18	19.0	1.056
hydrogen carbonate (aq)	CHO_3^+	61	140.3	2.300
carbon dioxide (g)	CO_2	44	94.2	2.141
carbon dioxide (aq)	CO_2	44	92.3	2.098
citrate	$C_6H_5O_7^{3-}$	189	278.7	1.475
creatine	$C_4H_9O_2N_3$	131	63.1	0.482
cysteine	$C_3H_7O_2NS$	121	81.2	0.671
carbon (c, graphite)	C_n	12n	0.0	0.000
chlorine	Cl_2	71	0.0	0.000
calcium (c)	Ca_n	40n	0.0	0.000
calcium (aq)	Ca^{2+}	40	132.2	3.305
calcium hydrogen phosphate	$CaHPO_4$	138	401.5	2.909
dihydroxyacetone phosphate	$C_3H_7O_6P$	170	308.9	1.817
erythrose 4-phosphate	$C_4H_9O_7P$	200	343.8	1.719
ethanol	C_2H_6O	46	43.4	0.943
formaldehyde	CH_2O	30	31.2	1.040

formic acid	CH_2O_2	46	85.1	1.850
formate (aq)	CHO_2^-	45	83.8	1.862
fructose	$C_6H_{12}O_6$	180	218.7	1.215
fructose 6-phosphate	$C_6H_{13}O_9P$	260	420.0	1.615
fructose bisphosphate	$C_6H_{14}O_{12}P_2$	340	621.3	1.827
fumarate	$C_4H_3O_4^-$	115	144.3	1.255
galactose	$C_6H_{12}O_6$	180	220.6	1.226
glucose	$C_6H_{12}O_6$	180	219.1	1.217
glucose 6-phosphate	$C_6H_{13}O_9P$	260	420.5	1.617
glutamate	$C_5H_8O_4N$	146	166.5	1.140
glutamine	$C_5H_{10}O_3N_2$	146	125.4	0.859
glycerol	$C_3H_8O_3$	92	116.7	1.268
glycerolphosphate	$C_3H_9O_6P$	172	319.2	1.856
glycine	$C_2H_5O_2N$	75	90.0	1.200
glyceraldehyde 3-phosphate	$C_3H_7O_6P$	170	307.1	1.806
hydroxly	HO^-	17	37.6	2.212
hydrogen (g)	H_2	2	0.0	0.000
hydronium ion (aq)	H_3O^+	19	56.7	2.984
hydrogen sulfide	H_2S	34	6.5	0.191
hydrogen cyanide	HCN	27	-28.7	-1.063
iron (c)	Fe_n	55.8n	0.0	0.000
iron(II) (aq)	Fe^{2+}	55.8	20.3	0.364
iron(III) (aq)	Fe^{3+}	55.8	2.5	0.045
isocitrate	$C_6H_5O_7^{3-}$	189	277.1	1.466
isoleucine	$C_6H_{12}O_2N$	131	82.2	0.627
α-ketoglutarate	$C_5H_4O_5^{2-}$	144	190.7	1.324
lactate	$C_3H_5O^{3-}$	89	123.4	1.387
lactose	$C_{12}H_{22}O_{11}$	332	362.0	1.090
leucine	$C_6H_{13}O_2N$	131	85.1	0.650
methane	CH_4	16	12.1	0.756
methanol	CH_4O	32	41.9	1.309
magnesium (c)	Mg_n	24.3n	0.0	0.000
magnesium ion (aq)	Mg^{2+}	24.3	109.0	4.486
nitrite ion (aq)	NO^{2-}	46	8.2	0.178
nitrate ion (aq)	NO^{3-}	62	26.4	0.426
nitrogen	N_2	28	0.0	0.000
oxalate	$C_2O_4^{2-}$	88	161.2	1.832
oxaloacetate	$C_4H_2O_5^{2-}$	130	190.4	1.465
oxygen	O_2	32	0.0	0.000
phosphoric acid	H_3PO_4	98	-274.1	-2.797
dihydrogen phosphate ion (aq)	$H_2PO_4^-$	97	271.2	2.825
hydrogen phosphate ion (aq)	HPO_4^{2-}	96	261.4	2.723
phosphate ion (aq)	PO_4^{3-}	95	245.1	2.580
phosphorus (c, white)	P_n	31n	0.0	0.000
pyruvate	$C_3H_3O_3^-$	87	113.4	1.303

phosphoenolpyruvate	$C_3H_5O_6P$	136	303.3	2.230
potassium (c)	K_n	39.1n	0.0	0.000
potassium ion (aq)	K^+	39.1	67.5	1.726
ribose (aq)	$C_5H_{10}O_5$	150	179.65	1.198
ribose 5-phosphate	$C_5H_{11}O_8P$	230	382.2	1.662
ribulose 5-phosphate	$C_5H_{11}O_8P$	230	381.7	1.660
quartz (c)	$(SiO_2)_n$	60n	192.4n	3.207
phenylalanine	$C_9H_{11}O_2N$	165	49.5	0.300
sedoheptulose 7-phosphate	$C_7H_{15}O_{10}P$	290	457.1	1.576
succinate	$C_4H_4O_4^{2-}$	116	164.9	1.422
succinyl CoA	$C_{25}H_{40}O_{20}N_7P_3S$	882	164.0	0.186
sucrose	$C_{12}H_{22}O_{11}$	342	370.7	1.084
sulfate ion (aq)	SO_4^{2-}	96	177.3	1.847
sulfite ion (aq)	SO_3^{2-}	80	118.8	1.485
sulfur (c, rhombic)	S_n	32n	0.0	0.000
silicon (c)	Si_n	28n	0.0	0.000
sodium (c)	Na_n	23n	0.0	0.000
sodium ion (aq)	Na^+	23	62.6	2.722
sodium chloride (c)	NaCl	58.5	91.8	1.569
serine	$C_3H_7O_3N$	105	122.1	1.163
threonine	$C_4H_9O_3N$	119	122.9	1.033
tyrosine	$C_9H_{11}O_3N$	181	92.5	0.511
tryptophan	$C_{11}H_{12}O_2N_2$	204	29.9	0.147
urea	CH_4ON_2	60	48.7	0.812
valine	$C_5H_{11}O_2N$	117	86.0	0.735
xanthine	$C_5H_5O_2N_4$	153	33.3	0.218
xylulose	$C_5H_{10}O_5$	150	178.7	1.191
water	H_2O	18	56.7	3.150

Notice how many important biological compounds are listed in the table. The qualifiers next to some of the names, (aq), (c) and (g) refer to the state of the substance which can be (aq)ueous, (c)rystaline or (g)aseous respectively. The strongest tendencies toward formation from the elements (*i. e.* the most positive entries in the table) occur for aqueous ions, quartz, calcium phosphate and water, however most entries have values between 0.5 and 2.0. There are simple sugars, amino acids, a purine (xanthine) and

numerous compounds that occur in metabolism in the table. Clearly, life evolved from those materials that were naturally occurring spontaneous products of the most abundant elements. Their formation from the elements is driven by a decrease in Gibbs free energy.

V. The monomer to polymer transition: no life to life

Life as we know it is distinguished by the presence of macromolecules: proteins, polysaccharides and polynucleotides. These provide structural elements, catalysts, energy storage, and information storage and processing components for the cell. The dynamical cellular **processes** inside the cell that we recognize as **living properties** are almost exclusively associated with these polymers.

Polymers are chains of monomeric constituents that have been joined together by covalent bonds. In the cases of proteins and polynucleotides, these chains are linear, whereas in the case of polysaccharides, they are branched. In all cases, the chemical structure of the linkages that join monomers into polymers is that of *dehydration condensates*. This means that the linkage between any two adjacent monomers anywhere in the chain can be broken by hydrolysis, *i. e.* cleavage by a molecule of water, literally "hydro-lysis". Most cells are 80-95% water which means that the tendency towards hydrolysis is strong, although perhaps not rapid unless catalysts are

available. An acid environment and digestive enzymes are needed by our stomachs to process food because the spontaneous rates of hydrolysis are too slow, but these processes are spontaneous because they are attended by a decrease in Gibbs free energy. Thus, the problem for organisms is the reverse, *i. e.* the synthesis of polymers from monomers, an uphill process thermodynamically that requires the input of Gibbs free energy. It is for this reason that the metabolic pathways are organized around the generation of energy rich compounds, particularly ATP (adenosine triphosphate) the almost universal currency of free energy for the cell.

The transition of matter from no life to life is in essence the transition from monomers to polymers. The monomers are formed spontaneously from the elements and have negative free energies of formation relative to the elements. The polymers, however, have positive free energies of formation relative to the monomers from which they are synthesized. Most would appear in the table around a value of 0.9 plus or minus 0.15. Making the "peptide" bond between amino acids in proteins costs about 2-3 kcal/mol, the "glycosidic" bond between sugar monomers in polysaccharides costs about 3-4 kcal/mol, and the "phosphodiester" bond between mononucleotides in polynucleotides costs around 5 kcal/mol. In each case, activation is essentially a phosphate activation, slightly disguised one way or

another. Indeed, for proteins the intermediate activated species is the aminoacyl-adenylate, a phosphate that is part of AMP (adenine monophosphate). The adenine of ATP can be substituted for by uracil, thymine, cytosine and guanine, generating the activated monomers UTP, TTP, CTP and GTP, from which, along with ATP, all DNA (deoxyribonucleic acid) and RNA (ribonucleic acid) chains can be made. In these substances, the sugar, ribose, occurs for the RNA case and the sugar, deoxyribose, occurs for the DNA case. When deoxyribose is involved the notation dATP, dGTP, dCTP and dTTP is used. The activation of sugar monomers involves UDP (uridine diphosphate) derivatives. Thus in all three cases, proteins, polysaccharides and polynucleotides, the activated monomers are phosphorylated compounds. A single phosphate is used for aminoacyl-adenylates that are AMP derivatives, a diphosphate is used for uridine diphosphate monosaccharides that are UDP derivatives and triphosphates are used for UTP, ATP, dATP, TTP, CTP, dCTP, GTP and dGTP that ultimately are all generated from ATP by phosphate transfers.

The required extra free energy is ultimately provided by ATP through formation of activated monomers, that once activated, spontaneously condense into polymers, but only with the aid of complex catalytic molecular machinery. Thus, dehydration and activation are universal

structural and synthetic themes for all life as we know it. Moreover, the use of energy rich phosphate compounds to achieve activation is also universal. The problem for the origin of life is how these phosphate compounds came into being before there were enzymes to catalyze energy metabolism. From an origin of life and energy perspective, this is the fundamental “chicken and egg” problem.

Elaborate macromolecular structures have been evolved that make activation possible in all extant cells. Many enzymes support a complex chain of oxidation reactions that harvests redox energy from high energy electrons. Not high energy in the physicist’s sense but high energy in the sense of biochemistry. The electrons may be excited initially by the intense UV irradiating the nascent Earth. Or reduced compounds with high electrical potentials (in Volts) can be generated in gentle thermal processes that dry out the reaction mixture. Drying means that water is leaving the system as steam and is taking with it the thermodynamic barrier to dehydration condensations. In the redox case, the electrons engage in a series of electron transfers, called redox reactions, that can couple to other energy requiring processes such as monomer activation.

The purest form of redox process is of the type exemplified by the ferrous-ferric transition

$$Fe^{2+} \Leftrightarrow Fe^{3+} + e^{-}$$

There are many instances of this type of redox step in the electron transport chains of bacteria and in mitochondria and chloroplasts, the energy transducing organelles of eukaryotic cells. Two major classes of Fe based transfers are found in all forms of life, those involving iron-sulfur proteins, and those involving cytochrome, a heme-Fe containing protein. These different species of Fe containing proteins contain individuals with redox potentials nearly spanning the range from free Fe (0.771 V) to ferredoxin-Fe (- 0.420 V). This permits evolution of a chain of many relatively small steps in the total decrease of Gibbs free energy, affording many opportunities for coupling to other processes.

The most rudimentary alternative to the simple electron transfers of Fe is co-transfer of a proton with an electron, *i. e.* a hydrogen atom in pieces. The ubiquitous ubiquinone species, found in all organisms possessing electron transport chains, are of this type and are responsible for the mechanism of chemiosmosis in membrane bound electron transport chain complexes. When the fundamental electron-proton, current-current coupling occurs in mitochondria, a decrease of Gibbs free energy during the electron transfer is partially used to increase Gibbs free energy for a steepening concentration gradient (that for a proton means a pH gradient), and an

increasing transmembrane electrical potential because the proton is positively charged. This generates the chemiosmotic “proton-motive force”, that is actually an electrical potential for protons (in Volts), not a force. Many processes, most notably transport of all sorts of molecules across the membrane, are driven by this form of cellular energy, rather than by ATP. Indeed, ATP is synthesized chemiosmotically in bacteria, mitochondria and chloroplasts by wonderful rotary enzyme complexes. ATP is ideally suited for activating monomers for the synthesis of polymers. However, there are some ATP driven ion transporters as well. Since chemiosmosis probably evolved after life began, phosphate energy was the universal currency of energy for the origin of life, but only with the aid of another sort of energy to be discussed in the next section.

The evolution of the cell membrane is unknown. Contemporary cellular membranes are primarily made from the class of lipids called glycerophospholipids that contain two fatty acid side chains and a phosphorylated alcohol such as choline, ethanolamine, inositol or serine. The fatty acid side chains contain between 14 and 24 carbon atoms. An elaborate enzyme complex catalyzes a long sequence of reactions in order to make these highly specialized lipids. Published models for an early evolution of a primitive precursor to these glycerophospholipids, in an

abiotic geochemical thermal process, make an earlier evolution of chemiosmosis more plausible.

Glycerophospholipids are needed to form lipid bilayer membranes. These structures, the essence of the cellular envelope, spontaneously self-assemble from aqueous solutions of lipid molecules. The more easily generated fatty acids, or single tailed lipids, produced by early Earth's geo-chemo-physical processes only form *micells* instead of the larger and more versatile bilayer *vesicles*. Did the cellular membrane structure evolve before life evolved, during the prebiotic chemical evolution phase? Or did it require a sophisticated metabolic machinery before its molecular constituents could be generated in sufficient quantities to comprise cells? Wouldn't such an apparatus necessarily have to be contained within a membranous boundary in order to keep enzyme and substrate together? This is another "chicken and egg" question.

Evidence suggests that before there was life as we know it, there were already morphological structures resembling cells, the bilayer vesicles, and there was a source of chemical energy through coupling to the electron transfer processes. These electron transfers run downhill spontaneously but through couplings to other processes, are generative of high energy chemicals. The oxidation of carbohydrates by molecular O_2 releases

enormous amounts of Gibbs free energy (almost 700 kcal/mol for one mole of glucose and six moles of O_2), about 40% of which is harvested by the pathways of glycolysis, generation of acetyl CoA, the citric acid cycle and the electron transport chain.

Carbohydrates are made by plants using the energy of sunlight, now in the visible part of the spectrum rather than in the primordial UV. Looking at table I, it is seen that carbohydrates, sugars, have less negative Gibbs free energies of formation than an equivalent amount of methane and carbon dioxide. The composition of many sugars can be written as a simple integer multiple, n, of formaldehyde, i. e. as $(CH_2O)_n$. Thus, the point is made by the fact that the process

$$CH_4 + CO_2 \rightarrow 2CH_2O$$

is attended by an increase in Gibbs Free energy of about 43.9 kcal/mol. It can be argued that sugar formed by whatever means can spontaneously convert back to methane and carbon dioxide by the inverse of the process shown above, a process referred to as carbon disproportionation. Since this releases energy, it has been argued that it is a possible source of prebiotic energy. O_2 is also made by plants. This means that glycolysis (that does not directly involve O_2) is more primitive than the citric acid cycle or the electron transport chain (that are directly linked to O_2). Interestingly,

glycolysis couples the carbon disproportionation of glucose to the generation of high energy phosphate in the form of ATP. The dimer, “pyrophosphate”, is the energy rich portion of ATP and is already sufficiently energy rich that it alone can drive synthetic processes such as polymerizations. Pyrophosphate, as well as other polyphosphates, can also be made abiotically by simply using gentle heating to dryness, or by coupling to thioester generation, as will be discussed in the next section.

There might well have been myriads of micron sized bilayer microspheres along with abundant reducing potential from high energy electrons and abundant pyrophosphate before there was true cellular polymer synthesis. This micron scale, energy rich environment appears ideally suited for the evolution of polymerization machinery. Only within such a simple energy rich environment can the required variety of polymers form at all.

Before true macromolecular polymers emerged, a smaller class of mixed oligomers, the coenzymes appear to have emerged. Coenzymes are the active catalytic moieties of many enzymes. They contain small molecular components that are called vitamins as well as ribose, phosphate and various other constituents, usually about half a dozen units of various kinds per coenzyme. The presence of phosphate, indeed pyrophosphate, and sulfur in many of them suggests that these species are relics of an early stage of

chemical evolution when energy transactions were already dominated by thioesters and phosphates. Since the coenzymes are thermodynamically uphill from their constituents, because they contain exclusively dehydration linkages, they could come into being only if the chemical milieu was one rich in energy transducing species. With their emergence, a rudimentary metabolism, still devoid of modern protein catalysts, could have also emerged. This chemical environment, perhaps housed in a membranous vesicle, would provide the setting for the ultimate development of polymer synthesis.

VI. Energy: redox reactions, thioesters, phosphate

The nature of life as we know it is dominated by the properties of polymers, most notably the proteins and the polynucleotides. They are responsible for the catalytic and informational characteristics of cells. For them to exist at all in an aqueous environment, Gibbs free energy is required to overcome the thermodynamic barrier to their synthesis. The energy for monomer activation, the prerequisite to polymer synthesis, is supplied by phosphate compounds. The concentration of inorganic phosphate in all bodies of water on the contemporary Earth is 10^{-5} times what it is in living cells. This makes it extremely unlikely that pyrophosphate was immediately available on the primitive Earth for the purpose of monomer activation.

Some more primitive, and abundant, supply of energy was needed first to couple the redox energy generated by UV and Fe to synthetic chemical energy. The solution to this puzzle appears to be the thioester.

The basic structure of a thioester is

$$R' - S \sim \overset{\overset{O}{\|}}{C} - R$$

where R' and R represent any of a great variety of molecular groups, or residues. The tilde denotes the high energy nature of the S and C bond. These energy rich compounds can form from carboxylic acids and thiols without the aid of organismic enzymes by either of two energy requiring mechanisms. The generic reaction is a dehydration reaction:

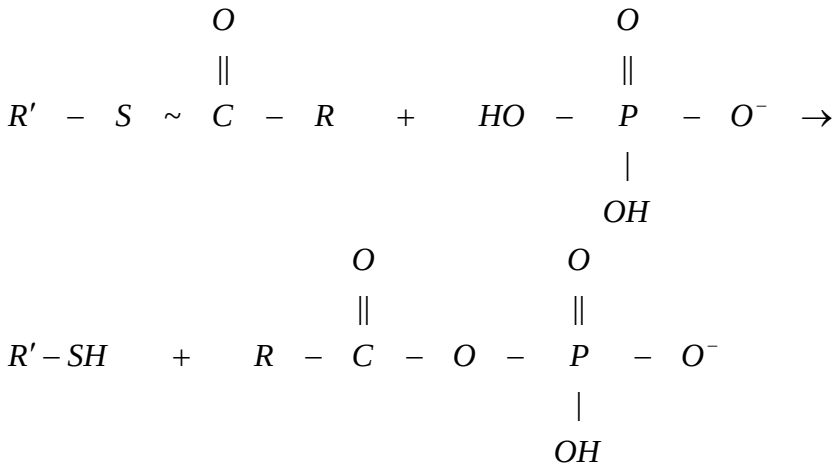
$$R'-SH + R - \overset{\overset{O}{\|}}{C} - OH \rightarrow R' - S \sim \overset{\overset{O}{\|}}{C} - R + H_2O$$

Variations on this theme in which the carboxylic acid on the left hand side is an aldehyde or an α-keto acid are also possible, in which cases two electrons and two protons, or two electrons, two protons and a carbon dioxide molecule are the byproducts respectively rather than a molecule of water. One of the two energy requiring mechanisms is simple heating of the thiol and the carboxylic acid at low pH. This promotes the dehydration. The other

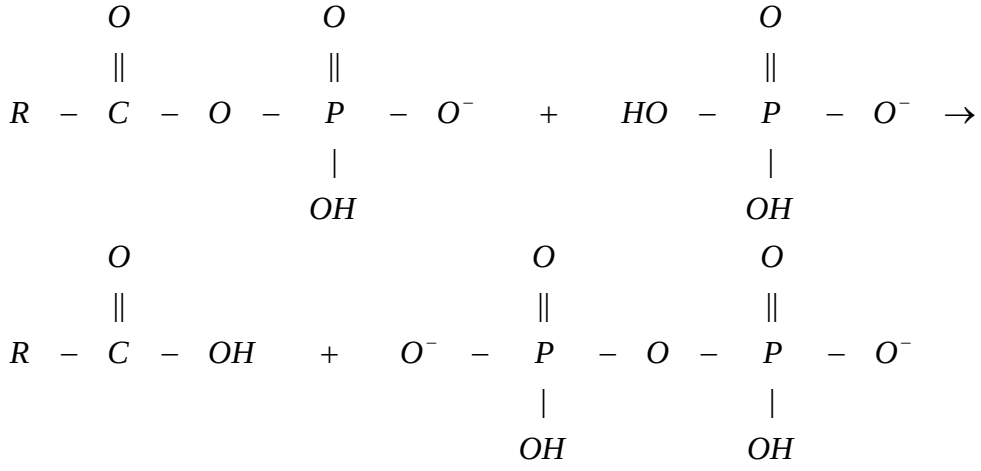
mechanism works well for an aldehyde and a thiol, or an α-keto acid and a thiol, wherein the oxidation by two Fe^{3+} ions takes away the freed electrons, leaving as byproducts the thioester and two protons, or two protons and a carbon dioxide molecule respectively. The ferric ions for this mechanism are the product of UV irradiation of Fe^{2+} as was discussed earlier. It is even possible at room temperature and neutral pH to spontaneously form thioesters from glyceraldehyde and a thiol.

Once formed, thioesters may be reduced to form reduced organic compounds not naturally produced abiotically. Reductive carboxylation to form dicarboxylic acids is also possible. Reductive amination of an α-keto acid will produce an α-amino acid. The electrons needed for these reductions, and many others, could come from Fe^{2+} irradiated by UV. These reactions represent precursors to many contemporary metabolic reactions that today are catalyzed by enzymes. In these modern mechanisms, the role of a thioester intermediate is very often retained in the active site of the enzyme. An example of this can be found in glycolysis.

In order to get phosphate into the picture the first step is the formation of acyl-phosphates. These form spontaneously through phosphorolytic attack of a thioester by inorganic phosphate



These acyl-phosphates are highly reactive and very energy rich. Acetyl-phosphate is an example of such a compound in contemporary metabolism. Inorganic phosphate attacks the acyl-phosphate to form pyrophosphate, a process that has also been shown to occur without enzymes.



Thus, the transduction of energy from redox energy, or heat, through thioester intermediates and into pyrophosphate is plausible on the primitive Earth before life *per se* had arisen. A large variety of molecules can form from the thioesters, and reducing potential generated from UV and Fe

oxidation, that are still biologically relevant today. Indeed, life as we know it is consistent with the proposition that it is these naturally occurring compounds that form the basis for life.

It is popular to refer to various stages of the evolution of life as "worlds", e. g. the "RNA world" to which we will return later. The earlier stage of chemical evolution described above can be called the "iron-thioester-pyrophosphate world." It represents the fundamental significance of energy transductions underlying the ultimate development of polymers, and the overwhelming importance in energy transductions of three elements, P, S and Fe. However, because P is so scarce, this world must have been preceded by the "iron-thioester world." This world lasted for some time and provided a mechanism for the slow but steady accumulation of phosphate. Once the "pyrophosphate world" emerged, really the iron-thioester-pyrophosphate world, the origin of coenzymes could leave its signatures.

VII. Banded iron formations in the geological record

Banded iron formations are world wide layered sedimentary deposits rich in iron that range in age from 1.5 billion years old to 3.8 billion years old, the age of the earliest known rocks. They contain between 30% to 60% ferric iron, Fe^{3+}. No molecular oxygen was present in the Earth's atmosphere until between 1.5 and 2.0 billion years ago. Thus the oxidant earlier could

not have been O_2. O_2 is now the ultimate oxidant for nearly all organisms but its function as such depends upon a complicated enzyme complex. By contrast the ability of UV to oxidize Fe^{2+} is geophysically natural and even autocatalytic. It has been demonstrated in the laboratory at acid, neutral and alkaline pH's. Since Fe^{3+} can serve as an oxidant for other molecules, Fe^{2+} can be regenerated. This creates an iron cycle. Depending on the stage of chemical evolution for all other molecular species, the amount of Fe^{3+} in any geological era could have gone through cycles as well, leading to the layers of magnetite minerals as the banded iron formations.

In an aqueous environment, the UV oxidation of Fe^{2+} forms Fe^{3+} and is followed by coprecipitation of ferric and ferrous oxides, Fe_2O_3 and FeO respectively. These oxides form from the iron ions and water molecules releasing protons as a byproduct as well as the mineral precipitate, magnetite.

$$2Fe^{3+} + Fe^{2+} + 4H_2O \rightarrow 8H^+ + [Fe_2O_3][FeO]$$

The released protons are available for the reduction of a large variety of substances and could have been responsible for the production of such species as H_2, CH_4, HCN and NH_3 as long ago as 3.8 billion years. These reduced species are otherwise not readily produced in other abiotic processes and are essential for the subsequent chemical evolution of amino acids and

nucleoside bases, the purines and pyrimidines. It should be noted that UV energy was one of the most abundant forms of energy on the primitive Earth.

Ferric iron, Fe^{3+}, may well have served as an early electron acceptor and, thereby, supported the oxidative synthesis of thioesters as was argued in the last section. Iron-sulfur proteins are among the most ancient known proteins and may be relics of an earlier iron-thioester-pyrophosphate world, but cleary not of the iron-thioester world.

VIII. The importance of d-orbitals

It is thought that the singular importance of P and S in energy transactions and the noteworthy unfitness of Si for metabolic chemistry is due to the properties of elements in the thrid period of the periodic table and also to d-orbitals. These electron states show up in the third period of the periodic table but are unoccupied in Si, P and S. Moving from left to right within a period, atoms actually get smaller even though they get larger as the period increases. Thus all atoms in the third period are bigger than those in the second period, but within the third period the size order is Si > P > S. Like C, N and O, P and S can form double bonds, but Si is just too big to do so too. Thus Si cannot form the vast variety of compounds that C does and is restricted to single bonds forcing it into immense three dimensional polymers of $(SiO_2)_n$. Moreover its unoccupied d-orbitals afford an

opportunity for attack of the relatively open Si-Si bonds by nucleophiles such as O_2, H_2O and NH_3. In contrast the bonds of P and S are tighter and their relatively smaller size allows them to form double bonds. The same d-orbitals that are such a liability for Si are instead an asset for P and S. For phosphates in particular, the result is a chemical group that is easily transferred while relatively impervious to chemical attack. In contrast, the smaller atoms of H, C, N and O produce very stable bonds that would not be suitable for easily transferred groups. Thus, phosphate appears to be a natural choice for the activation group in synthetic biochemistry. The d-orbitals make it pentavalent and this contributes to the resonance enhancement of its energy transactions that confer on it its “high energy” status.

IX. Thermal energy and synthesis

Thermal energy has been mentioned briefly so far. It is of central importance for the origin of life in two distinct ways. At the molecular level it gives rise to a very robust energetic activity called Brownian motion. The collisions between water molecules and other molecules are very energetic and are responsible for the Brownian motion of the other molecules. This Brownian motion makes proteins fluctuate vigorously in conformation and promotes the catalytic activities of these molecules. Indeed, without the

Brownian motion, the protein enzymes would lose their functionality. Brownian motion is also responsible for very rapid mixing of species within a micron sized environment. The other importance for thermal energy is that it can overcome the thermodynamic barrier to dehydration polymerizations. Since this barrier is a result of the overwhelming presence of water molecules and the fact that all polymer linkages are dehydration bonds, thermal energy, *i. e.* heat, can remove the water molecules if applied in a gentle enough manner. Too much heating will simply destroy the molecules and create a gooey, tarry mess. For origin of life considerations, it is this second significance for thermal energy that is of synthetic importance.

A prime example of the synthetic power of thermal energy is the thermal synthesis of polyamino acids, called thermal proteinoids. This type of polymer synthesis works especially well if the amino acid mixture is rich in aspartic acid and glutamic acid, not unlike the composition of many contemporary biological proteins. It is also assisted by the presence of phosphate, that by itself yields polyphosphates when heated. Probably the polyphosphates formed in the amino acid mixture help promote condensation of the amino acids themselves. Such phosphate enhanced reactions mixtures yield thermal proteinoids at temperatures as low as 60^{o} C if heating is allowed for say 150 hours. It is also of interest to note that when

the thermal proteinoids are returned to water after synthesis, they spontaneously form uniform populations of micron diameter microspheres. Since many amino acid residues are lipophilic, these thermal polymers have some properties in common with true glycerophospholipids. In particular the proteinoid microspheres are bilayered vesicles rather than micells. The proteinoids also possess a number of relatively weak catalytic abilities covering the basic reaction steps that make up metabolism. All of this together strongly suggests a natural mechanism for the formation of catalytic microphere environments in which the iron-thioester-pyrophosphate world could gain a foothold. While the proteinoid microphere membranes are not nearly as impermeable as the membranes of modern organisms, they would nevertheless trap any polymers formed within them from monomers that could freely enter them from the environment. With the evolution of a more sophisticated metabolism, incorporation of primitive lipid analogues would make the membranes much less permeable and permit the establishment of chemiosmotic mechanisms. The proteinoid model has received a great deal of criticism in the literature. However, a close study of the primary literature presenting the experimental results that support the model clearly refutes much of the criticism.

X. The RNA world

Since the discovery of ribozymes, RNA molecules with enzymatic activity, in the early 1980's, it has been very popular to speak of the origin of life in terms of the RNA world. The basic notion is that RNA simultaneously has the ability to serve as the genetic material and as the first enzymes. Thus, the answer to the question: "which came first, proteins or polynucleotides?" receives the emphatic answer, polynucleotides, specifically RNA. This question is an old chicken and egg question and opinion has alternated between proteins first and polynucleotides first for some years. At present, a more evenly balanced view seems warranted given the difficulties surrounding the chemical evolution of mononucleotides, and especially their important ingredient, the ribose molecule, that so far is not easily produced in abiotic experiments. In earlier sections of this article, it has been clearly established that the spontaneous production of polymers is thermodynamically inhibited. RNA polymers require phosphodiester linkages, the dehydration linkages for polynucleotides, and these are uphill in Gibbs free energy. This situation is doubly a problem for polynucleotides because they are made from monomers, the mononucleotides, that are themselves oligomers, *i. e.* very small polymers. ATP, for example, contains adenine, ribose and three phosphates. Each component is linked to its neighbors by dehydration linkages, for a total of four in the case of ATP or

any other mononucleotide triphosphate. Thus, even the oligomeric monomers for polynucleotide synthesis are uphill in Gibbs free energy relative to their constituents. Given these thermodynamic barriers, coupled to the difficulty so far in explaining an easy natural origin for ribose, it is highly unlikely that an RNA world was the beginning stage for life as we know it. For this reason, the RNA world section of this article is the last section, not the first.

In the preceding sections it has been argued that an energy transducing chemical evolution preceded the true emergence of life. Before the RNA world could have emerged, the iron-thioester-pyrophosphate world had to be in place, perhaps already encapsulated inside microspheres with proteinoid and/or lipid precursor membranes. Recall that phosphate, an essential component of polynucleotide phosphodiester backbones, is scarce on the Earth and only through the aid of thioesters does it seem likely that it was recruited from the geophysical environment into the biological realm as was described in section VI. If it is asked: which came first, the RNA world or the iron-thioester-pyrophosphate world?, then it should now be clear that it was the latter.

Once the essential energy requirements for the transition from monomers to polymers are established, the RNA world becomes possible. It

this sense, the origin of life, the transition from monomers to polymers, coincides with the emergence of the RNA world. The problem then shifts from one of origins to one of evolution. The key issue is that of protein biosynthesis by a gene directed mechanism. The evolution of this mechanism must account for the emergence of ribosomes, tRNA's (transfer RNA), mRNA (messenger RNA), rRNA (ribosomal RNA) and the aaRS's (aminoacyl-tRNA synthetases). The aaRS's of contemporary organisms are a diverse and complex group of enzymes responsible for attaching the cognate amino acid to a transfer RNA with a specific anticodon. The mechanism of this recognition process in contemporary organisms is far from completely understood and all of the structural and evolutionary evidence so far strongly points to a long evolution of the mechanism and its components. This unsolved problem remains the big gap in our comprehension of the contemporary genetic mechanism.

Models for how these complex components could have evolved from much more primitive precursors have been proposed. They purport to explain: 1) why the genetic code is a three base code with lots of degeneracy in the third codon and why the codons for the amino acids are what they are; 2) why the N-terminus to C-terminus direction of the gene directed protein is colinear with the $5' \rightarrow 3'$ direction of the cognate mRNA; 3) why L-amino

acids and D-ribose are used, *i. e.* whether this is a relative relationship that could as easily have been D-amino acids with L-ribose instead, or it is an absolute necessity; and 4) how gene directed synthesis of proteins got started before there was the complex array of protein and RNA catalysts that currently make the process work. There are many other related issues. One published model proposes that the first genes and mRNA's were one and the same molecule, an RNA, and that protein synthesis took place directly on this RNA when acitivated amino acids interacted directly with the $2'-OH$ groups of the ribose moieties to form amino acyl ribose esters. Through a subsequent conformation change the esterified amino acids link up to form small proteins and release the RNA for another round of synthesis. The RNA has served as both gene and messenger in this model. The activated amino acids might be carboxyl phosphates generated from pyrophosphate or they might be thioesters. Subsequent evolution of DNA, lacking the $2'-OH$ groups, would separate the roles of information storage in DNA and information translation in the RNA. This very simple genetic system would be poised for the subsequent evolution of ribosomes, tRNA's and aaRS's. Such models provide conceptual insight into origins and evolution, and also provide a number of challenges for experimentalists. However, recent

advances in biotechnology make possible a number of experiments testing the ideas just presented.

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