Chemical evolution and the origin of life

I. INTRODUCTION

Evolution appears to be a fundamental feature of matter. It is an observed general tendency of matter to change quantitatively its degree of structural complexity as a function of time. One speaks therefore of the evolution of the stars, of the earth, of the living organisms of molecules, and atoms.

The subject of this study is that part of evolution which has led, on the earth, from simple molecules to distinctly living organisms.

The very nature of our subject, the origin of life, excluded the possibility of writing about it in a rigourous scientific way (i.e. by dealing only with reproducible observations or experimental facts). We will nevertheless try to keep our speculations within the scope of modern science by using for the construction of the ideas under discussion whenever possible only well established facts and by trying to bring these ideas to a point where some kind of experiment or observation can help us to evaluate their soundness.

Before we engage in speculations about the origin of life there are some general questions we have to answer first.

1. *Has life an origin* and if so *did it originate on earth*?

To answer this question we can use several lines of argument.

a. The Biological argument: wherever the evolutionary lines of animal or plant species are known, they show a tendency to converge; if we extrapolate from this observation we come to the conclusion that all these organisms must be descendants of only one or a few primitive organisms and that therefore life probably had an origin in such primitive organisms. This argumentation was in fact already implicit in DARWIN's and WALLACE's concept of the origin of species. According to their concept new species originate from the different varieties formed by a previously existing species. Extrapolation from this concept also points to an origin of life.

b. The Geological and Geophysical arguments:
It is generally agreed that conditions on earth have once been such that life, as we know it, was impossible on the surface of the earth. Furthermore very old rockformations are known in which no trace of life has been found up to now.

c. A general argument:
If we accept that the concept of evolution is in fact a general property of all matter whereby a gradual change of complexity of structure occurs, we must view the living organisms as a stage in this general evolution and hence they must have had an origin in time.

From arguments a and b we can safely conclude that life on earth must have had an origin.

The question can now be asked whether life itself has an origin or whether it was always present in the universe. Argument (c) points to an origin, but gives no answer to the question whether this origin took place on the earth or somewhere else in the Universe and was subsequently inoculated onto the earth when conditions became favourable on its surface.

At present the evidence for life outside the earth is scarse and even if it turns out that some other planets support life it would still be possible that this life had an independent origin. We will therefore assume in this discussion that life originated on earth.

2. *What is life?*

It is obvious that before we can set out to discuss in what possible ways life originated, we should have a clear knowledge of what life or a living organism is in comparison with non-living matter. The usual way to summarize this knowledge is to give a sharp and specific definition of a living organism which would completely set it apart, with no possibility of confusion, from all non-living structures. It turns out however that this is quite impossible. Many definitions of life have been given stressing one or another or many typical characteristics of living organisms but not one of them allows one to draw a sharp and distinct line between living and non-living matter.

All the materials out of which living organisms are built invariably also form part of clearly non-living structures. (Both the living and non-living world are built from the same elements).

All the typical and essential features of living organisms such as: self-reproduction; irritability; structure movement, anabolism and catabolism etc., taken separately are not exclusively found in living organisms. Moreover there is no evidence that the atoms and molecules of living organisms would obey physical and chemical laws different from those known to rule the behaviour of the atoms and molecules of non-living structures. (cfr. Vis Vitalis) Nevertheless, as Schrödinger points out, it is unlikely that it will be possible to reduce the way in which living matter is working to the ordinary laws of physics, but this is probably only due to the fact that we are dealing here with an altogether peculiar degree of complexity and a special kind of energy exchange.

According to Schrödinger the most striking difference between living organisms and non-living matter is that the first are able to accumulate « negative entropy » or « order » thereby escaping, temporarily and at the expence of their environment, the general tendency of the universe to increase its entropy and thereby its atomic disorder (2^o law of thermodynamics). However systems accumulating negative entropy or building up an orderly structure at the expence of their environment are also known in non-living systems (e.g. formation of crystals).

The difference therefore is mainly in the rate and the extent to which this temporary building up of order by increasing the disorder of the environment, takes place.

From the above it can be concluded that it is not possible to give a sharp definition of life.

This, of course, could be due to lack of knowledge, but it is more likely, in fact, that the absence of a sharp and absolute boundary between non-living and living matter is due to the very nature of life itself and that, as knowledge increases, this boundary will become less and less sharp.

The best way therefore to answer the question : « what is life » is to give a description of the essential features common to all living organisms.

The first of these features is « *structural organisation* ». All the components of living organisms are put or lay embedded in a very definite structural order usually called a cell.

The actual composition and degree of complexity of these cells can vary from organism to organism but it remains true that all living organisms consist of one or several of these structural elements.

That an ordered structure is essential for life can also be concluded from the fact that all the main constituents of living cells show an amazing degree of order. This is not only true for such visible structures as nuclei, mitochondria, chromosomes, membranes, centrosomes etc. but also for the proteins, lipids, mucopeptides, nucleic acids and in general for all the big molecules found in living cells. It has been one of the finest achievements of present molecular biology to show that nearly all the constituents of cells down to the molecular level are very highly structured and minutely ordered. A second feature is that all living cells are built up from essentially the *same type of molecules* and polymers consisting mainly of only a small number of different elements. Hydrogen, carbon, oxygen, nitrogen and phosphorus account for the formation of the bulk of all living matter. These elements form amino acids, sugars, purine and pyrimidine bases and acids and these molecules in turn form highly structured polymers such as proteins, polysaccharides nucleic acids and lipids which when put in a very specific order account for the formation of a very wide variety of living cells.

A third general feature is the capacity of all living organisms *to maintain* with a very high degree of efficiency *their specific order and structure.* This they achieve by a system of self-reproduction whereby living cells continuously synthesize their specific molecules so that they will grow and divide. The most striking fact here is that this reproduction is very specific. A given cell will, except for relatively rare mutations, always produce cells practically identical to itself over a very large number of generations. Summarizing we can say that life is the result of a high degree of structural order and complexity in an association of polymers of proteins, nucleic acids, polysaccharides and lipids forming cells endowed with an efficient system for self-reduplication. We therefore conventionally call living, systems having a certain minimum degree of structural complexity such as found in a cell.

If we want to solve the problem of the origin of life we shall thus have to look for a development giving rise to these kinds of polymers and to this kind of order starting from non-living physical elements (without this type of self-reproducing ordered complexity).

Indeed it follows from what has been said in the preceding pages that we must see the origin of life as a result of and as a stage in the general evolution of matter. Prior to the origin of living organisms there must have been chemical evolution giving origin to the complex polymers specific for living organisms. It is this evolution that we shall try to describe in the following paragraphs and subsequently the possible ways in which these polymers got organised in order to form a living cell.

II. Theoretical aspects of the origin and the evolution of life.

Living organisms are open systems maintaining their structural and functional integrity by a constant flow of energy, which is degraded in the process. The ultimate source of this energy is to be found in the sun, where irreversible reactions take place, enabling the sun to radiate energy into the universe. By this radiation the sun gets rid of the entropy increase (1) which accompanies the irreversible energy yielding reactions and thus allows the sun to maintain a steady state. This steady state enables the sun to produce constant radiation, part of which reaches the earth, from where it is re-radiated into outer space at a lower energy level. In other words, the earth is an open system receiving energy from the sun and re-radiating this energy at a lower energy level, thus increasing the entropy of the universe. The net result of the combined activities of the sun and of the earth is an increase of the entropy of the universe, which is in agreement with the second law of thermodynamics. This law can be stated as follows : for each transformation in an isolated system, the entropy of the final state is never less than the entropy of the intial state. Hence it follows immediately that the state with maximal entropy is also the most stable state, as each change from this maximum would imply a decrease of entropy, which would contradict the second law. This law can give information bearing upon the direction of processes taking place in isolated systems : if these systems change, they will evolve in the direction of higher entropy.

We already pointed to the fact that living organisms are open systems but by no means isolated ones. It thus appears that the second law of thermodynamics cannot be applied here in its original form. Prigogyne however has made extensive studies of the thermodynamics of open systems, and the results of his calculations appear to be of great value in understanding the evolution of life.

An open system is characterized by the uptake of matter and/or energy out of its environment, the transformation of it within the system, and the release of the transformed products into the environment.

fig. 1

$$A \rightleftarrows (A \rightleftarrows B) \rightleftarrows B$$

If an environment containing a substance A (fig 1) can be considered infinite, the system represented can never reach thermodynamic equilibrium but remains in a steady state of dynamic equilibrium. The composition of the open system in its steady state depends only on the reaction rate in the system. Fluctuations of the concentrations of A and B will

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(1) Entropy : the several exact definitions of entropy fall beyond the scope of this article, For readers not aquainted with this notion it wil be sufficient to state that, in a most general way, the entropy measures the disorder present in a system.

cause changes in the reaction rates such as to restore the original steady state. Such a simple open system thus shows a certain degree of what might be called homeostasis. But there is more: RASHEVSKY was able to show that the forces generated by the diffusion of A and B tend to disrupt the open system when its volume attains a critical value. Under certain conditions, namely when a substance is produced, an open system which is smaller than the critical volume will grow and will divide upon reaching the critical volume. Each daughter system will first restore the original reaction rate, will grow and will later divide in its turn. This provides us with an elementary picture of growth and multiplication. So we see that very simple open systems share some properties with the more complex open systems we call living, indeed they show some form of self regulation, growth and multiplication.

The work of PRIGOGYNE also bears upon the evolution of these open systems. He was able to show on thermodynamical grounds, that open systems will evolve in the direction of increasing complexity, moving towards greater thermodynamical efficiency: minimal production of entropy by irreversible stationary processes. Different open systems can compete with each other, and the calculations show that those systems with the highest efficiency will survive. If different paths are possible, open systems will evolve towards systems with the highest reaction rates. As these reaction rates depend upon the presence and the nature of the catalysts within the system, and as we already saw that the organisation of open systems is dependant only on these same reaction rates, it is clear that the evolution of open systems will be determined by the evolution of the catalysts within them. In a certain way the story of life is the story of the evolution of catalysts and enzymes. But these open systems are still far from reaching the complexity of living cells. The question arises whether there are some theoretical reasons to think that this primitive open systems should evolve in the direction of cellular organisms.

We saw that the general direction of the evolution of open systems was towards increasing complexity. But there is an upper limit to this complexity: the kinetics of biochemical reactions depending only on random collision of the reactants in a fluid medium do not allow the size of the system to exceed 1 micron, if an optimal reaction rate is to be maintained. It thus appears that in their evolution towards complexity open systems will reach a point where the thermodynamical efficiency, which is the driving force of their evolution, will be impaired unless there is a simultaneous evolution of structural elements. Once these structural elements are present, the reactions can take place in an orderly way, not any longer being dependant on random collisions and open systems with an internal morphological structure will be able to supplant more primitive open systems.

This is in good agreement with the calculations of RASHEVSKY who showed that, according to the theory of algorithms, structural complexity is accompanied by functional simplicity. If this principle holds in the biological world, and there is some evidence that it does, we have here another reason to expect open systems to evolve some structural order.

As a corollary of the evolution towards complexity it appears that the need arises for a more exact replication mechanism in the open system. It is clear that spontaneous division of an open system will only produce two daughter systems which are more or less alike if the system is not too complex. RASHEVSKY was able to show that no matter how a simple open system is divided and how its content is stirred, the diffusion gradient will be automatically re-established. This is no longer the case with complex open systems where we expect some mechanism assuring an equal distribution of the principal constituents between the two daughter systems. Probably a mechanism assuring that the different catalysts present within the open system were equally divided between the daughter systems would suffice, as the organisation of the open systems depends only on these catalysts. Furthermore, if this system is to have what we now call genetic properties, its stability should be such that it could exist during a time exceeding the life time of the open system of which the genetic mechanism is part.

Concluding we may say that, given a source of energy (the sun) an environment (the universe) into which entropy can be given off and the existence of simple open systems, these systems, which already show homeostasy, growth and multiplication, will evolve towards greater complexity.

This evolution will be dominated by the evolution of the catalysts in the open systems. Upon reaching a certain degree of complexity further evolution will become impossible unless the evolving systems acquire morphological complexity enabling them to maintain a high thermodynamical efficiency. The multiplication of such systems will become impossible when they reach such a degree of complexity that spontaneous division will not be sufficient any longer to ensure accurate reproduction. At this point the existence of an exact replicating mechanism becomes more and more a necessity.

It thus appears that the problem of the origin and the evolution of life can be reduced to the question : how did open systems with their enzymes and genetic mechanism originate.

A. The Origin of open systems

We saw in the foregoing part that the development of the structures we call living can be seen as the result of the evolution of simple open systems. This supposes that organic substances which we think were formed abiogenically were assembled into one system. The simplest means by which this could have happened is by the formation of coacervates. By coacervation we understand the formation in a colloid solution of two separate phases: a colloid-rich phase, the coacervate, and a colloid poor phase, the equilibrium liquid. The coacervate separates out from the liquid as a continuous layer or, more frequently, as tiny droplets, floating in the liquid. In the classic experiments of Bungenberg de Jong with protein colloids these droplets has a diameter ranging from 2 to 670 micron. These droplets have a definite internal structure owing to the rigid association of the molecules within them. This same association of the molecules provides also a sharp boundary limiting the droplet. Furthermore the concentration of proteins within such coacervates is in general much higher than in the colloid before coacervation occured, and a fortiori much higher than in the liquid phase. The proces of coacervation thus provides us with a mechanism for obtaining a high concentration of organic and inorganic products enclosed in a small volume which is isolated from the environment. If some reaction took place in such coacervate droplets then these droplets could be the primitive open systems postulated in the theoretical part.

The ability to form coacervates is widespread: it occurs in many hydrophylic and hydrophobic colloids, both organic and inorganic. It is however of interest to note that proteins easely form coacervates, because these protein coacervates could be the primitive systems we suppose to be the origin of living systems, which nowadays are composed chiefly of proteins.

If at some stage of the evolution of these protein coacervates, lipids became an integral part of them, this would mean the beginning of the structural complexity of the system. For it is known that lipids associate readily with proteins to form membranes having a high degree of structural stability. These lipoprotein complexes are of utmost importance for living cells, where they form the many membranes and organelles which are responsible for the orderliness of the reactions taking place in the cell.

We think it not unreasonable to assume that the common phenomenon of coacervation gave rise to the simple open systems which may be the origin of life. The origin of the proteins needed for building the simplest coacervates and the origin of the lipids needed for the morphological organisation of the more complex open systems therefore appear to be the main questions we have to answer.

B. THE ORIGIN AND EVOLUTION OF ENZYMES

In the theoretical introduction we have shown that the evolution of simple open systems depends in the first place on evolution towards increased efficiency of the catalysts present in them.

The catalysts of present-day cells, the enzymes, are very highly ordered and usually very specific complex protein molecules, the synthesis of which requires the complicated organisation of a living cell.

These enzymes must be the result of a very long process of evolution, involving gradual improvement from very much simpler initial systems, derived from the original simple solution in which the abiogenic chemical evolution towards polymers must have started. In fact these systems must have had a chemical composition not too different from the composition of the primitive solution.

Since probably the main energy yielding reactions taking place in the primitive coacervate droplets were redox reactions (processes involving the transfer of hydrogen) it is relevant to examine the possible evolution of compounds catalysing this type of reaction as an example. It is worth pointing out that reactions of this type are the most common reactions in the metabolism of all organisms. Many simple compounds present in the primitive solution such as metal ions, have catalytic functions of this type. For instance the hydrated ferric ion has a peroxidase catalytic activity of a very low order. (In this reaction hydrogen peroxide is decomposed to water and oxygen).

If, however, this iron is surrounded by a suitable organic grouping — such as a tetrapyrole (forming a haem) — it now has a catalytic activity one thousand times as great. If this haem forms a still more complex structure by combining with a protein and thus forming the enzyme « catalase » the catalytic activity is increased by another factor of about a million.

This example shows that the catalytic power of a simple catalyst such as iron can be tremendously increased by building it into a more complex structure. We can thus imagine that among simple open system e.g. catalysing the decomposition of hydrogen peroxide into water and oxygen, there was great selective advantage for those open systems where by a random process, the iron was incorporated into a more complex and also more active structure.

There must have been an evolutionary trend leading to open systems having catalase enzymes starting from simple ones having iron as a catalyst. We think this example of the abiogenic evolution of a simple catalyst to an enzyme can probably be applied to most primitive enzymes.

Another factor of great importance in the evolution of enzymes is the phenomenon of *auto-catalysis.*

Consider the sequence of reactions A → B → C → D ····· → X leading to the random synthesis of a complex catalyst. If this complex catalyst can itself catalyse any of the reactions in the sequence its production will be enhanced.

For example, let us suppose that step B → C is catalysed by a simple catalyst such as Fe^{++} and the end-product X is the molecule haem. If haem is a better catalyst for this reaction than iron itself then there will be auto-selection for those steps catalysed by haem and this will lead to a more efficient and more regular formation of haem. Open systems possessing this sequence of reactions will then have a selective advantage over those that do not.

In future it may be possible to explain the evolutionary origin of most of the basic enzymes starting from simple catalysts present in the primitive solution according to the following principles:

1. the existence of simple open systems depends primarily on the catalysts in them.
2. the catalytic activity increases dramatically when simple catalysts are incorporated into more complex structures and combine with proteins thus forming enzymes.
3. that auto-catalysis provides a way to build into open systems a sequence of reactions by which the more complex catalysts can be synthesized more regularly and are gradually less and less dependant on random synthesis for their formation.
4. competition between simple open systems for the necessary substrates in the primitive solution provides a powerful selection mechanism as a result of which more efficient and therefore more complex open systems will evolve.

It remains to be shown that some of these more complex catalysts and especially proteins can be synthesized randomly in the primitive solution. This we will do in section III.

C. The origin of a Genetic mechanism

It is an inherent property of open systems to grow and to divide but as the structural and chemical complexity of such systems increased, the necessity for accurate partitioning of the various components at each division must have become more and more important. As the chemical and structural complexity increased the chances that even an equatorial division would bring about an equal partitioning of all the essential components of the system would decrease. What in essence is required is some sort of short hand summary of all the essentials of the system in a chemically and structurally compact form of such a nature that replication can be achieved easely and accurately. What do we know in nature that fullfills this role?

The accumulated evidence of the last decade points unambiguously to nucleic acid as the material with all of the features demanded. It is now beyond reasonable doubt that nucleic acids are the chemical basis of heredity. The evidence upon which this assertion is based is manifold. First there was the demonstration by *Avery*, *Mac Leod* and *Mc Carthy*, that the active principle mediating genetic transformation in bacteria was DEOXYRIBONUCLEIC ACID (DNA).

The capacity of DNA to transmit hereditary properties from one strain of bacteria to another is completely and specifically destroyed by DNA-ase an enzyme which destroys DNA only. The greater the degree of purity of the DNA employed in this genetic transformation experiments, the higher ist specific activity. To agree that DNA is not the chemical base of heredity demands that we assign a quite untenable amount of genetic information to the very small amount of contaminating protein present in the very best preparation of transforming DNA.

The second line of evidence comes from a study of the life of bacterial viruses. *Hershey* and *Chase* showed that upon infection of sensitive hosts all the virus DNA enters the host cell while almost all of the protein remains on the outside and can be removed without affecting the further development of the virus. Thus we can say that the virus DNA programmes the whole of the development of the new virus particles.

Plat viruses provide us with further evidence. Tabacco mosaic virus (TMV) consists of ribonucleic acid and protein in the form of a rod of RNA surrounded by a coil of proteins. These two components can be separated physically and when this is done it is found that the protein fraction is inert while the RNA fraction alone is infective and can direct the formation of whole new TMV particles.

All of this evidence comes from the rapidly developing field of microbial genetics and it is not unreasonable to ask whether the evidence, presented so far, represents a few, admittedly well studied, but nevertheless special cases? Similar studies using rapidly growing cultures of human tissue cells instead of cultures of bacteria have been initiated and there are reports that transformation of some of the properties of these cells can be accomplished using preparations of DNA obtained from genetically distinct lines.

Less rigourous but nonetheless important is the fact that nucleic acids are an essential component of the nuclei of all the known cells and are localised in the chromosomes. Chromosomes are visible structures in the cell nuclei and it has been known for many years that they are the physical seat of the genetic information of both animal and plant cells. The mode of their division is well documented and the genetic traits they determine are partitioned in exactly the same way.

Furthermore there is the fact that the somatic cells of any given organism contain a very constant amount of DNA irrespective of the structural or functional differentiation of the cell or its metabolic state, whereas all other components of such cells are subject to enormous variations. Haploid cells contain only half the amount of DNA present in diploid cells.

Finally those agents, both physical and chemical, which have the most direct effect on the genetic behaviour of cells can be shown to act directly on nucleic acids.

From all this and other evidence : it can safely be concluded that nucleic acids are the chemical basis of heredity and since nucleic acids have been found in all living organisms it is very probable that all the living organisms have the same basic genetic mechanism. Some recent experiments concerning the biosynthesis of proteins support this view. This is very important because if all living organisms have the same genetic mechanism it is reasonable to assume that also the most primitive of them had this mechanism and hence it is for nucleic acids we have to look if we want to understand the origin of genetic mechanisms.

What then are the remarkable characteristics thanks to which nucleic acids are able to perform this essential function? First it must be capable of rapid and precise replication. Secondly it must have inherent in it the capacity to change (mutate) and so provide a base upon which selection can be exercised and thirdly it must be capable of storing information in a compact form and to yield this information rapidly and efficiently where this is requested. We shall show that recent studies on the chemical structure of DNA illustrates that this substance does in fact possess these most remarkable characteristics.

Physico-chemical structure of DNA

Nucleic acids are composed of three essential components, a pentose sugar, phosphoric acid and nitrogeneous organic bases. The nitreogeneous bases are of two kinds: purines and pyrimidines. In DNA the purines are ADENINE (A) and Guanine (G) and the pyrimidines Thymine (T) and Cytosine (C), while in RNA Thymine (T) is replaced by Uracil (U).

The component formed by combination of the sugar, base and phosphate groups is called a nucleotide and thus in DNA there are four types of nucleotides depending on the four bases. Chemical analyses have revealed that DNA is a polymer built up of long chains of nucleotides joined by sugar-phosphate links in a repeating pattern. Almost invariably there is an equivalence of purine and pyrimidine nucleotides (i.e. $A + G = T + C$) and moreover there is equivalence of A and T and of G and C. However this does not mean that DNA from all sources has the same composition. In fact the base ratio $A + T / G + C$ varies from 0.35 to 2.07. X-ray diffraction analysis has given further insight into the structure of DNA. By this method repeating patterns of molecules can be detected in a meso crystal.

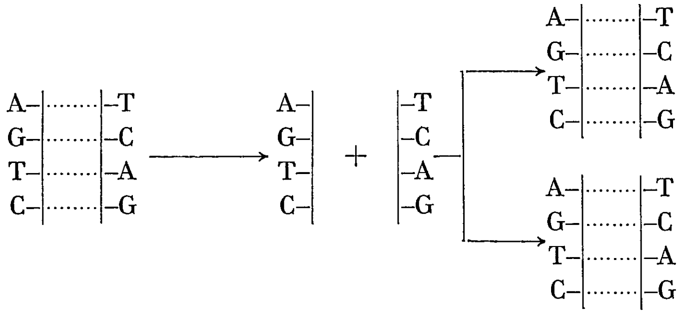
It was shown that

a) the bases are arranged at right angles to the polymer chain
b) the chain is not straight but is a regular helix
c) there are two chains in each helix.

Watson and *Crick* brilliantly synthesised the available information into a precise model which was not only compatible with all the available data but which possessed just those properties which are demanded of DNA if it is to serve as the chemical basis of heredity i.e. replication, mutability and to act as information store. This model of DNA comprises two polynucleotide chains which are complementary to each other in the sense that A in one chain is always placed opposite T in the other chain and G is always placed opposite C. The two complementary chains are wound round each other to form a double helix.

Let us now consider in what way this model fullfills the requirements of genetic material:

1. *The genetic material replicates*: since the two strands of DNA are complementary to each other exact replication is accomplished by strand separation, followed by the synthesis of new single strands, each of the old strands serving as a template for the synthesis of a complementary one:



2. *The genetic material must act as an information store*

In the Watson and Crick model this information lies in the pattern of the helix. There is no restriction in any one chain on the sequence of the four bases and the information is coded in the sequence of the bases in the chain. According to this hypothesis the sequence of the four bases in the DNA determines the sequence of amino-acids in the enzyme proteins made by the cell. (One can compare this to the alphabet where the sequence of letters forming words and sentences convey an information. The genetic code would then be a four letter alphabet).

3. *The genetic material can undergo mutations to provide a basis for genetic variability and thus for evolution*

According to the Watson-Crick model mutations would consist of alterations in the pattern of base sequences of the DNA replicates and would be revealed by alterations to the proteins coded for by the DNA and thus provide an altered phenotype on which selection can be exercised.

Experimental evidence and consequences of the Watson-Crick model of DNA

It is beyond the scope of this article to quote in any detail the experimental evidence in support of the Watson-Crick model of DNA.

Experimental evidence in favour of this model is accumulating every day coming, from different lines of approach and obtained with different kinds of organisms. It is therefore safe to state that this model is amply documented and generally agreed upon. This holds also true for the genetic code. Specific alterations to the DNA have been experimentally induced by chemical treatment and the subsequent changes in particular proteins have been identified and found to consist of particular amino-acid substitutions.

The transcription mechanism

One can now proceed to answer the question as to how a particular sequence of bases in DNA can be transcribed in order to form a particular sequence of amino-acids in a protein. From the coding DNA a complementary single strand of RNA is formed thus having a complementary sequence of bases in it. This transcribed single stranded RNA is called « Messenger RNA ». These messenger-RNA molecules shift from the nucleus, where they are formed, to some particular structures in the cytoplasm of the cell (the ribosomes). Here they serve as a template for the synthesis of the proteins. To each specific sequence of probably three bases in the messenger-RNA an other type of RNA (Transfer RNA) gets attached (there are at least as many different transfer-RNA's present in the cell as there are different amino-acids participating in the formation of proteins). One can see that this way a particular sequence of bases in the messenger-RNA will have a particular sequence of transfer-RNA's with specific amino-acids, attached to it. The adjacent amino-acids are subsequently linked together by chemical bonds and the specific protein polymer thus formed is detached from the messenger-RNA, transfer-RNA complex.

The regulation mechanism

In a living cell not all the genes are operating all of the time. The cells request for different enzyme functions varies both in quantity and in time. Thus regulation of enzyme synthesis is an essential request. How is this regulation accomplished?

It has been found that special regions of the DNA act as switches which determine whether a sequence of bases is read or not (thus indirectly whether a particular enzyme is synthesized or not). This switch is turned of by small molecules called repressors which are most probably allosteric proteins with a dual specificity : one for the operator and one for a metabolite.

When a particular metabolite is present in excess all the repressor molecules for a particular operator will be charged and the operator will be switched off. This mechanism acts essentially as a feedback control. There is accumulating evidence that DNA synthesis itself is under a similar form of control.

The origin of the genetic mechanism

In the foregoing we have discussed the need for a genetic mechanism (to achieve a perfect partitioning of structurally complex cells) and the mechanism itself. We have shown that the nucleic-acids of the chromosomes are able of replicate, to carry genetic informations and to mutate. The purine and pyrimidine bases are arranged in a specific sequence along the DNA molecule and this linear sequence of bases determines, thanks to a special transcription mechanism, the linear sequence of amino-acids in proteins. The proteins themselves are responsible for all the properties of the cell since they form both the structural backbone and enzymes of it. It is very important to note that, as for as we know, essentially the same genetic mechanism functions in all living organisms. It is therefore justified to suppose that the most primitive living organisms had a genetic mechanism in which the orderly structure of nucleic acids had an influence on the structure and the order of amino-acids in proteins. In order to understand the origin of a genetic mechanism we will therefore first have to look for the origin of nucleic acids. (See Section III).

III. Some experiments and observations in relation to the origin of life

A. Introduction

The theoretical considerations of the previous section reveal that the problem of the origin of life can be reduced to the *origin of open systems, enzymes and a genetic mechanism.* In fact the principal problem is the abiogenic origin of proteins, nucleic acids and lipids since these are the main constituents of membranes, enzymes and the chromosomes. To investigate this problem we need some knowledge about the conditions which prevailed at the time when chemical evolution was taking place. The consensus of opinion today is that organic molecules were first formed in the primitive hydrosphere and atmosphere surrounding the earth and that the energy required for their synthesis was derived from solar radiation.

It thus becomes necessary to determine as far as possible the composition of the primitive hydrosphere and atmosphere and to try to deduce what chemical reactions could take place in them under influence of solar radiation. The most remarkable feature of the earth's present-day atmosphere is its content of free oxygen. It is the only planet so far on which free oxygen has been detected. How can we account for its presence?

Several independent isotopic dating methods indicate that the solar system was formed about 5×10^9 years ago. It is very probable that the first atmosphere of the earth was similar to that in the cloud of dust and gas from which the solar system was formed and would thus consist mainly of hydrogen and helium. It can be readily calculated from the formulae of Jeans that the light elements H and He would be very quickly lost into interstellar space. But as a result of a series of physical and radiochemical processes this atmosphere was replaced by a secondary one consisting mainly of CH_4, H_2, CO_2, N_2 and some NH_3, CO_2 and H_2S. The lighter gases CH_4 and H_2 were gradually lost and the gases CO, CO_2 and N_2 increased in proportional concentration. The solubility of NH_3, H_2S and CO_2 reduced their concentrations in the atmosphere as they combined with elements in the earth's crust leaving essentially an atmosphere of nitrogen. The accumulation of free oxygen was hindered by its combination wich elements in the earth's crust with which it formed oxides and it did not constitute an appreciable fraction of the atmosphere until biogenic processes began.

The following table summarises the early development of the earth's atmosphere.

Stage of development

	I	II	III	IV
	Formation	Heating and condensation	Transition	Biogenic
	Reducing	Reducing	Reducing	Oxygenic
Main components	H_2, He	CH_4, H_2 CO, N_2	N_2, CO_2 CO	N_2, O_2
Lesser components	N_2	CO_2 rare gases soluble gases	CH_4 rare gases	CO_2 rare gases

It can be argued that the oxygen in the atmosphere of the earth was derived from purely chemical and physical processes during its early formation. What cannot be denied is that its persistence is due to biogenic processes. The mass of the earth and its attendant force of gravity is insufficient to prevent the escape into space of free oxygen so it must be replenished. The only significant source of replenishment is the process of photosynthesis.

To determine the time at which this interesting change in the atmosphere took place, i.e. from a basically reducing condition to an oxygenic condition representing a change from abiogenic to biogenic processes we have to resort to geological evidence.

The normal geological sequence of changes in rock at the earth's surface is weathering, erosion, transportation and sedimentation. Weathering may be physical, due to extremes of temperature, or chemical. The type of chemical weathering which occured is determined by the composition of the atmosphere in contact with the rock. So if we could determine the type of chemical weathering which occured in deposits of known age we could determine the nature of the atmosphere at the time of their formation.

Under oxygenic conditions chemical weathering leads to the attack of all minerals except quartz and ore oxides, such as magnetite, which are left unchanged. The products of chemical weathering are in general soluble and during transportation and sedimentation further chemical changes take place leading to the production of clays. This then leads to two types of sedimentary deposits : quartz sands and clays. Limestone deposits are largely of biogenic origin.

Under reducing conditions the sequence of events will have been different. There felspars, the so-called dark minerals (biotite hornblende etc.) and sulphides will not have been oxidized. They could have been involved in repeated cycles of erosion, transportation and sedimentation without being chemically altered, and subject only to physical and mechanical stresses. Repeated cycles of this sort would lead to efficient sorting by size and weight of the particles in the deposit. Under these conditions sands containing minerals other than quartz would be formed and would consist of various particle sizes. In fact the physical rather than the chemical composition of the sediments would be the main factor in determining their structure.

We shall consider two lines of geological evidence bearing on the nature of the early atmosphere. The first comes from a study by *Rankama*, who has attempted to evaluate the character of the early atmosphere from the chemical composition of the sediments formed during the relevant period. He studied the detailed deposits around a granite rock from which they were most probably formed. The granite rock contains 4% ferros and 2% ferric iron. Under oxidative weathering the relative amount of ferric iron will increase and should exceed the amount of ferrous iron. This was not so. In fact, the proportion of ferric to ferrous iron was even less than in the original granite. Thus it is reasonable to conclude that this granite weathered under reducing conditions. A tentative age of 2×10^9 years has been assigned to the deposits and so we can say that a reducing atmosphere certainly persisted at that period.

The second piece of geological evidence bearing on the nature of the early atmosphere comes from studies by *Randohr* on the pre-Cambrian deposits of the Old Shields of S. Africa, Brazil and Canada. These deposits are formed of ancient sands and gravels. They represent sediments laid down by repeated cycles of weathering and erosion and contain in addition to quartz, sulphides and pitchblende (derived from earlier uraninite, UO_2, the least oxidized of the uranium oxides). All these factors indicate formation under reducing conditions. Further the composition of all of these deposits is strikingly similar in spite of their world-wide geographical distribution. The detailed structure of the deposits shows the type of sorting by size and weight of the different components in the sediment expected when only physical and mechanical forces operate in their formation. In contrast the newer gravels and sands consist exclusively of quartz. The age of the deposits studied by Randohr is 2-3 × 10^9 years which agrees with that estimated by Rankama for the age of the reducing atmosphere.

A word of caution is necessary in considering this kind of evidence critically. First, in the type of study undertaken by Rankama it is obviously very important to ascertain that the sediment analysed is in fact derived from the rock with which its composition is compared. In the type of study undertaken by Randohr it is important to assess the importance of secondary processes such as redeposition which may affect the primary structure of a deposit.

From the above it is clear that a study of properly preserved old deposits yields information about the state of the atmosphere at the time of their formation and enables us to say that the early reducing atmosphere was still present $\pm$ 2 × 10^9 years ago. At what period did it become oxygenic? An answer to this question has been obtained from a study of the so-called red-beds which consist chiefly of quartz and acquire their colour from the small percentage of iron they contain. Quartz is stable under either reducing or oxidising atmospheric conditions, but iron is not. The iron in the red-beds is present in the highly oxidized ferric form as limonite $Fe_2O_3nH_2O$. Thus the red-beds must have been formed under oxygenic conditions. The oldest known red-beds are a little less than 1 × 10^9 years so by this time the atmosphere was already oxygenic.

We thus arrive at the period 1-2 × 10^9 years ago as the time during which the atmosphere changed from reducing to oxygenic. It is in this important transition stage that evolution developed from abiogenic to biogenic processes. The absence of free oxygen from the primaeval atmosphere has a very important consequence. It means that there would have been no shielding layer of ozone in the upper layers and therefore the short wave-length ultra-violet radiation from the sun could freely penetrate to the surface of the earth. Radiation of such short wave-length is of very high energy and can excite some free elements to form chemical bonds through the adsorption of light quanta. Under these circumstances typically organic molecules can be built up from the simple constituents of the primaeval atmosphere. This process has been called inorganic photosynthesis, and the compounds built up in this way would be relatively stable in an oxygen-free atmosphere.

Experimental confirmation of this process has been obtained by *Miller*. He showed that under the influence of short wave-length ultra-violet radiation organic compounds were produced by the formation of covalent bonds between H, CH_4, NH_3 and H_2O, in the complete absence of free oxygen. The first compounds produced appear to be HCN and aldehydes and later these decrease in amount as a series of amino-acids are synthesised. *Wilson* has experimentally produced much larger molecules under similar conditions. These molecules appear to be polymers of carbon compounds in the form of thin sheets.

More recent studies have shown that, given amino-acids, polymerisation to form long chain polypeptides can occur under conditions similar to those on the primitive earth. Not only have polypeptides (hence proteins) been produced in this way but also purines and pyrimidines have been synthesised from HCN in aqueous ammonia.

These recent experimental findings make it clear that under atmospheric conditions which have been established to exist on the primitive earth at the relevant time and using the materials and energy available at that time covalent bonds of organic compounds could be built up into quite complex molecules, including those compounds necessary for the initiation of biogenic processes.

IV. Summary and a proposal for an evolutionary origin of living cells

In order to have a living organism, one needs an highly structured organisation which has the ability to metabolize, to grow and to multiply actively at the expence of the environment. In the preceding chapters we have shown that such complex organisations can theoretically evolve from simple open systems if they acquire efficient catalysts and a mechanism for accurate replication.

The catalysts involved in living cells are called enzymes and are made of long chains of amino-acids forming protein macromolecules. These enzymes direct all the chemical and energy changes taking place in the cell which are responsible for its growth and multiplication.

The accurate replication of the cells is achieved thanks to a mechanism whereby directions for the synthesis of proteins are stored in the nucleic-acids present in the chromosomes of the cells. Thanks to a complex reading mechanism the linear sequence of bases in the nucleic acids is translated into a linear sequence of amino acids forming specific proteins. Furthermore in a cell the nucleic acid macromolecules are able to make very accurate copy's of themselves thus transferring their information from one generation to the next one.

The problem of the origin of living cells thus resolves itself into the following points:

1. The origin of simple open systems
2. The origin of enzymes
3. The origin of nucleic acids and a genetic code
4. The problem of the integration of these different elements into a functional unit: the living cell.

In sections II and III we have presented evidence supporting the idea that the various macromolecules, e.g. lipids, proteins and nucleic acids, originated by a purely chemical evolution under abiogenic conditions and that these macromolecules therefore existed before living cells emerged. It can safely be assumed that under the prevailing primitive conditions the simple elements present on the surface of the earth and in the atmosphere underwent a chemical evolution towards increasing complexity leading to the formation of membranes, coarcervates, proteins and nucleic acids. The main forces directing this evolution were the sun (mainly by its high energy ultra violet rays) and a selection, exerciced by the fact that in simple open systems structural complexity is accompanied by higher efficiency in the competition for the available building elements of which the macromolecules are composed.

The main problem thus becomes: how did these different macromolecules evolve and integrate in order to form a functional living cell?

To construct an hypothesis concerning this problem we shall again follow a reasoning based upon the *concept of natural selection.*

For better understanding of the following paragraph the reader is referred to section II (p. 17).

Usually the problem is stated in the following way: what came first, the proteins (enzymes) or the nucleic acids (the genetic mechanism)? It seems to us that this is a wrong approach and that both systems originated simultaneously and, in fact, evolved due to *mutual interactions.*

As long as both proteins and nucleic acids were formed independently by random chemical evolution, there was no way to preserve the more succesfull molecules, since these macromolecules have a limited life and the chances of repeatedly making the same molecules by a random mechanism are very low. Therefore a system allowing the repeated formation of a special type of macromolecule would have had a direct selective advantage. Moreover, if certain macromolecules are regularly formed they can constitute a basis for gradual and minute changes out of which selection would pick the more efficient ones, thus allowing an evolution towards increasingly efficient molecules of the kind we know in living cells.

Using the existing knowledge about the genetic mechanism (see section II C) and applying to this the foregoing reasoning, we can construct the following hypothetical model. By normal chemical reactions short protein and nucleic acid molecules were formed first. Among these, some small RNA molecules would have had the property of binding amino-acids and thereby activating them.

The system, small RNA-amino-acids, would then form protein molecules more readily and more efficiently than the simple chemical linking together of non activated or free amino-acids. At the beginning this binding by small RNA's of amino-acid molecules was probably not specific nor efficient. It is enough to suppose that some small RNA molecules had a certain affinity for some or any amino-acid, to have a beginning point for a gradual evolution towards « transfer »-RNA molecules with a high degree of specificity and affinity. The next step would be the formation of bigger RNA molecules (cfr. Messenger RNA) on to which the « transfer »-RNA molecules would get attached by base-pairing. The selective advantage of this system is two-fold : 1) in this way several transfer-RNA molecules with the activated amino-acids attached to them, would be brought in close linear contact with one another. 2) this « messenger » RNA molecule would serve as a kind of template onto which several more or less identical protein molecules could be formed.

Again there is no need to assume a very specific linkage between primitive « transfer »-RNA's and primitive « messenger »-RNA's. However small the degree of specificity in the beginning, this is nevertheless enough for an evolution toward increased specificity and efficiency.

The third step in this evolution would have been the origin of a mechanism whereby « messenger » or « template »-RNA could be preserved and accurately reproduced. Indeed these longer-RNA molecules are unstable and have a relatively short life-time. Therefore a mechanism whereby they would be regularly and accurately reproduced would have a high selective advantage. The more stable, dubble-stranded DNA molecules could have provided such a mechanism. In fact it is known that DNA molecules now function as a template for the formation of messenger-RNA by a mechanism whereby the two strands of DNA separate and form either new DNA strands or produce single stranded RNA copy's.

Here again things could have started with a non-specific mechanism serving as a base for selection and evolution towards a specific and efficiently working genetic mechanism.

As a result of, and all along this evolution starting from simple non-specific « transfer »-RNA's over « messenger-RNA's towards a complete genetic mechanism using stable and accurately replicating DNA-molecules, protein molecules of an increasing complexity and catalytic activity will have been formed, and in fact were the very point at which natural selection could be exercised since these proteins formed improved catalysts for the reactions taking place in the primitive open systems.

If one assumes that some of these primitive enzyme-proteins played a role in the formation first of « transfer »-RNA and later of « messenger »-RNA and double stranded DNA we can visualize the way in which proteins and nucleic acids made the primitive open systems evolve into living cells thanks to mutual interactions.

In conclusion it can be said that the basic idea of this paper is that living cells are the result mainly of the action of natural selection on such chemicals as proteins and nucleic acids.

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