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Asymmetry in Biomolecules an Evolutionary Overview

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ASYMMETRY IN BIOMOLECULES: AN EVOLUTIONARY OVERVIEW (*)

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Abstract. This review presents the biomolecules within the framework of the theory of evolution, after it has been extrapolated from the population level down to the cellular and molecular levels. From the theory of common descent - we may assume that probably all the organic beings have descended from a primordial cell (Darwin, 1859), a protocyanobacterium, but the last common ancestor may also be called a 'progenote' (Woese, 1983). This view has gained wide acceptance. Earlier still, prior to the encapsulation of nucleic acids in lipid microspheres, evolution may already have been at work at the nucleic acid level, on RNA molecules (the 'RNA world').The topics from the biophysics of the biomolecules that we have chosen to include are: the possible role of extraterrestrial amino acids in the origin of life, the origin of translation, which illustrates the intimate relationship between proteins and DNA through the standard genetic code. Finally we also refer to the origin of the lipids. Our main aim has been to demonstrate that proteins, together with nucleic acids and lipids, the three biomolecules that are emphasized during the Mini-symposium, are all intimately related to both structure and function and, consequently, are central topics for further research in biophysics.

1. Introduction

1.1. ON ASYMMETRY, BIOMOLECULES AND EVOLUTION

In spite of the fact that these are questions that underly modern science it may be useful to define the scope and limitation of these terms, all of which are intimately related.The concept of symmetry is a deep one in mathematics, the physical and natural sciences. Weyl, for instance stated that (Weyl, 1969):

> "If I am not mistaken, the word *symmetry* is used in our everyday language with two meanings. In the one sense symmetric means something like well-proportioned, well-balanced, and symmetry denotes that sort of concordance of several parts by which they integrate as a whole...The image of the balance provides a natural link to the second sense in which the word symmetry is used in modern times: *bilateral symmetry*, the symmetry between left and right, which is so conspicuous in the strtucture of the higher animals, especially the human body".

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Asymmetry, on the other hand, is a reflection of another well-defined concept, namely that of symmetry-breaking. This is best illustrated with the example of a buckled table-

tennis ball (Stewart and Golubitsky, 1992). Being originally a perfect sphere, it was invariant under the group of three-dimensional motions that fix its centre, the underlying group is the orthogonal group O(3) in three dimensions.

The distorsion introduced in the ball defines a new axis and the invariance is reduced to all the rotations through any angle about the axis, and reflexions in planes that contain that axis; in other words the two-dimensional orthogonal group O(2). This gives us a precise formulation to symmetry-breaking, namely a change of the symmetry group to another one of smaller dimensionality. Group theory, particularly the irreducible representations of the symmetry groups allows to address most relevant questions pertinent to the main subject addressed in this Mini-symposium.

The bilateral symmetry, referred to by Weyl, in the above citation, the symmetry between left and right, in the structure of the higher animals (except for the cnidarians), may have some reason behind it. Also the lack of such symmetry may have a causal factor. Both of these questions may be addressed at all levels, even at the level of molecules from the point of view of the theory of evolution of Darwin. But in particular, not all molecules can be part of living organisms.

The once mysterious question *What is life?* (Schrodinger, 1944) no longer retains the flavour of mystery that surrounded it when theoretical physicists addressed it in war-time Dublin. With the progress of molecular biology it may be seen that practical definitions may be assigned to the once elusive concept of life. Indeed we may propose that life is intricately associatecd with evolution. This concept lies at the basis of a working definition adopted by the Exobiology Programme of the National Aeronautics and Space Agency (NASA):

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life is a self-sustained chemical system capable of

undergoing Darwinian evolution
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(Coveney and Highfiels, 1995).

Regarding the more perinent questions as to which are the molecules of life, referred to in this Mini-symposium as the *biomolecules,* the question will be clarified in the course of the present lecture, as there is an enormous quantity of work done in the field of chemical evolution, since the pioneering work of the group of Melvin Calvin in 1951, in which a simulated atmosphere of water vapour, carbon dioxide and molecular hydrogen were shown to lead to the synthesis of formic acid, glycolic acid and, most significantly, formaldehyde one of the precursors in the synthesis of ribose (prominent in the initiation of the RNA world), and glycerol (prominent in the next fundamental step in the ascent of life, the generation of the cellular membranes and walls), cf., Table 3 below.

The theory of evolution of Darwin in fact includes two distinct aspects: the theory of common descent as well as that of natural selection (two distinct theories!). The theory of common descent that will be most useful to us in our determination to underline the intimate relationship that exists between all the biomolecules. Indeed, we have made an explicit effort to present a wide enough panorama in the Mini-symposium, not just a presentation of protein biochemistry and biophysics, possibly the most popular topic at present amongst biophysicists.

For this reason we now turn to the question of the origin of the biomolecules in terms of plausible chemical and physical processes occurring in an environment analogous to the terrestrial planets. In physical terms the problem can be stated as follows: As time may be introduced in an unambiguous way in Darwinian evolution by appealing to the protracted rate of mutation in all the organisms, DNA mutates at a steady rate in over geological time.

A related evolutionary clock may be controlled with the evidence of the available fossils in order to have a rational basis for taxonomy. The zeroth of this evolutionary time corresponds to the existence of the last common ancestor - the first cell - a topic that takes us into the problem of the origin and evolution of life in the universe.

1.2. THE ORIGIN OF THE BIOMOLECULES

From what we have said above the present review introduces the biomolecules in the context of the origin and evolution of the first cell. We extend some ideas first introduced iearlier (Chela-Flores, 1994b). These research topics are inextricably linked with a more basic question, namely the different scenarios for the origin of the first biomolecules.

Present-day organisms have inherited well defined asymmeric biomolecular relics through a mixture of contingency and consequences of natural laws. Some of the scenarios for the evolution of life are:

1.2.1. *The organizing template scenario.* The current process of translation may be thought of as having been originally based on an organizing template, which assembled the biomolecules. Such mineral substrates may have preceded the current genetic processes that code for amino acids (Cairns Smith, 1985). This conjecture is plausible since in the particular case of clay surfaces the substrate itself may have catalytic properties (Ferris *et al.*, 1988).

1.2.2. *The metallic mineral surface scenario.* This consists of one iron and two sulfur molecules (pyrite FeS_2, Wächtershäuser, 1990). He suggested that life may have begun as a purely metabolic process based on pyrite as a substrate.

1.2.3. *The protein-first scenario.* Primitive life may have consisted purely of metabolic machinery without replication. This alternative is a protein-before-nucleic acid scenario (Dyson, 1985).

1.2.4. *The proteinoid scenario.* The first proteins may have replicated directly; nucleic acids came later. Fox and co-workers have supported this view by synthesizing "proteinoids" (i.e., protein-like polymers) by means of heating amino acids (Fox *et al.*, 1995).

1.2.5. *The RNA world.* RNA came before DNA. About 4 billion years ago RNA may have preceded the present DNA-protein stage. This aspect of the origin of life- referred to as the "RNA world"- has been covered in recent reviews (Gestland & Atkins, 1993). We have discussed possible relics (Chela-Flores, 1995), the viroids (Diener, 1989) that the RNA world may have left in contemporary biota. This may lead to a paradox, in view of the known paleontological data. However, such paradox may have been clarified recently (Chela-Flores, 1994a; 1996).

1.2.6. *The Eigen-Schuster scenario.* Replication and translation may have coevolved (Eigen & Winkler-Oswatitsch, 1992; Schuster, 1993).

1.3. POSSIBLE ROLES OF ASYMMETRY IN BIOLOGY

If the origin and evolution of life on Earth was not entirely the product of contingency, one possible way to approach the outstanding questions of the origin of the main molecules may be with the help of symmetries: A symmetry in the laws of nature is a statement that implies that under certain changes in the point of view from which we observe nature a given law does not change (Weinberg, 1993).

Due to the nature of natural selection and the strong component of contingency, it is not common to view phenomena of the living world from the point of view of symmetries. Broken symmetries, in particular, underlie cooperative phenomena, which may be the key to homochirality, instead of the accumulation of frozen accidents. In general the role of broken symmetries is well understood in the properties of inert macroscopic matter including, for example, the cases of superconductivity and ferromagnetism (Anderson, 1972).

In condensed matter there is the possibility of cooperative effects. We have argued that cooperative phenomena also underlies living matter as well (Chela-Flores, 1985, 1987). It seems evident, as we propose to review in the present work, that a further stage in our discussion should be a property that goes beyond physics, namely information, separating the biomolecules from more orthodox forms of condensed matter.

1.4. EXTRATERRESTRIAL ORIGIN OF THE BIOMOLECULES ?

We should recall that the origin of life may be widespread in the universe, as the 35 year-old search initiated by Frank Drake has attempted to demonstrate (Drake and Sobel, 1992) This possibility is gaining support and is prominent in the forthcoming space missions: the Cassini probe of the European Space Agency (ESA), the Russian 1996 Mars mission, the ESA Rosseta Mission due to be launched early next century.

The eventual success of our enquiry may depend at the beginning ot next century on the adoption of truly significant enterprises, such as the extension of the search for extraterrestrial life (SETI) to the far side of the Moon (in the 100 km diameter crater Saha, as advocated by Jean Heidmann. None of the extremely ambitious engeneering projects underlying this proposal, such as a 340 km road linking laboratories in Mare Smythii and crater Saha, seem beyond present capabilities of humankind (Heidmann, 1996).

Against this forward-looking attempt to come to an understanding of our origins corresponding theoretical ideas elucidating the basis of chemical evolution should not be discarded given some present experimental difficulties (clearly, if profound theoretical reasons are put forward, these proposals should be discarded).

For instance, the critical temperature T_c in the Salam proposal (to be discussed in our contribution to the Mini-symposium) may be in principle too small for the phase transition to have been supported by the environment of the early Earth. Hence, the extraterrestrial option (now a reasonable proposition from what we have said above) should not be ruled out.

A further illustration of the plausibility of an extraterrestrial origin of the precursors of the biomolecules is based on the Murchison meteorite. On September 21, 1969, this meteorite fell in Murchison, Australia. The laboratory of Cyril Ponnamperuma was able to obtain a piece of it. At that time they were preparing for the first analysis of the lunar rocks.

They were able to get the first conclusive evidence of extraterrestrial amino acids (Ponnamperuma, 1995). The result of this analysis is shown in Table 1.

TABLE 1: Amino acids from chemical evolution experiments and from an extraterrestrial source (Orgel, 1994). The relative abundances of the amino acids in the chemical evolution experiments that imitate the paleoatmosphere as a mixture of CH_4, NH_3, H_2O and H_2. In the second column we show the amino acids found in the Murchinson meteorite (Knevolden *et al.*, 1971; Orgel, 1994; Wolman *et al.*, 1972). The relative amounts are denoted by dots.

Amino acid	*Murchison meteorite*	*Chemical evolution experiments*
Glycine	••••	••••
Alanine	••••	••••
Valine	•••	••
Proline	•••	•
Aspartic acid	•••	•••
Glutamic acid	•••	••

However, it should be remarked that the question of which were the gases that were present in the early Earth atmosphere is not settled (Kasting, 1993). For instance, carbon dioxide must have been abundant to have prevented the Archean Earth from freezing under the Sun at a lower level in the Main Sequence of the Hertzprung-Russell diagram (some 30% less luminous at the time of the origin of life-the 'faint young Sun paradox'). Through a greenhouse effect there would have ben the appropriate temperatures for producing the microflora that we know must have occurred some 3.5 Gy bp (Schopf, 1993).

1.5. CHEMICAL EVOLUTION

The question of reproducing the effect of the primordial atmosphere in the laboratory has been approached gradually. First of all, as briefly mentioned in Sec. 1.1 above, Melvin Calvin and co-workers in 1951 assumed the existence of a primitive atmosphere of CO_2, H_2O and H_2, which were constrined inside a closed flask (Garrison *et al.*, 1951). A source of energy was applied to this model of the paleoatmosphere; in fact, they used the Berkeley 60-in cyclotron. In this manner the organic products obtained were formadelhyde which is particularly relevant for life's origins (cf., Table 3 in Sec. 4.2), formic acid and glycolic acid (both of which are prominent in living organisms.

Later on, the model of the paleoatmosphere was enriched with the addition of methane (which is now known to exist in the atmosphere of Titan) and ammonia, after the effect of an energy source by Stanley Miller (Miller, 1953), the organic products included the amino acids.

Sydney Fox succeeded to produce protenoids by thermal heat in presumed primitive paleoatmospheres (Fox and Harada, 1958). The synthesis of the monomers of nucleic acids soon followed, notably adenine by John Oro (1961) from hydrogen cyanide in water-ammonia systems under prebiotic conditions, and independently, by Cyril

Ponnamperuma and co-workers from electron irradiation in a model of the paleoatmosphere (Ponnamperuma *et al.*, 1963).

1.6. WHAT IS HOMOCHIRALITY ?

This broad knowledge of early life has allowed a search for relics that may show the common thread of evolution from our last common ancestor. At the beginning of the 1990s Abdus Salam's deep interest in the role of asymmetry in the living state gave a strong impulse to the topic of the asymmetry of the biomolecules (Salam, 1991, Chela-Flores, 1991).

In all of these scenarios the first biomolecules exhibit a common feature, namely molecular chirality. This property is shared by DNA and proteins, as well as some of the components of the plasma membrane, as for instance the lecithins (Wald, 1957).

Chirality, in the restricted case of protein amino acids, is the first topic we shall discuss. Another possibility, however, should be kept in mind, namely that the effective complexity of the universe receives only a small contribution from the fundamental laws. The rest may come from numerous regularities resulting from frozen accidents, chance events of which the particular outcomes have long-term consequences all related by their common ancestry (Gell-Mann, 1994). In this view, the evolution of complexity in biosystems may be the accumulation of frozen accidents, or in other words (Gould, 1989) biocomplexity may be the product of contingency, i.e., the shaping of the present biota may result from a long chain of unpredictable antecedent states, rather than by the determination of the invariant laws of nature.

The first unifying principle in biochemistry (Crick, 1981) is that the key biomolecules have the same handedness: this phenomenon occurs when molecules are asymmetric, in such a manner that they are able to exist in two configurations, both partners ('isomers') being mirror images of each other. These two possibilities are referred to, according to the response of a mixture of them to the incidence of a plane-polarized light. More precisely, the angle α through which the plane of polarization is rotated can be given a sign as follows: when looking towards the incoming beam of light, for a clockwise rotation of α, we would assign the term 'right-handed', or dextrorotatory (D); if such a rotation is anticlockwise we say that the rotation is 'left-handed', or laevorotatory (L).

Molecules that respond to beams of light in the above-mentioned manner are said to be *optically active*. Examples of single-handed biomolecules that will concern us in this review are the monomers of proteins, which are only L-amino acids, while the ribose and 2-deoxyribose monomers of the nucleic acids are all in the D-configuration. Phospholipids are no exception. These are biomolecules of the cell membrane having a phosphate group and one or more fatty acids. Indeed, the plasma membrane of the organisms of the domains Bacteria and Eukarya contain exclusively D-glycerol (Kandler, 1995).

Thus, the key biomolecules have the same handedness, or, we say more often that biomolecules have the same 'chirality' or, preferably that they are 'homochiral'. (These words are taken from the Greek language, as *cheir* means hand.) Finally, an equimolar mixture of both isomers is called a 'racemic mixture'. Chiral molecules have non-superposable D, or L three-dimensional mirror image structures or 'enantiomers' (once again, these are words derived from the Greek *enantios morphe*, whose meaning is 'opposite shape').

It is important to distinguish between the truly chiral influences (such as the electroweak interaction), from other physical effects that have been proposed since Pasteur's famous resolution of tartaric acid crystals. (These are crystalline naturally occurring carboxylic acids, with melting point of 170 ^{o}C, which Pasteur showed to be optically active). In fact, Barron defines a truly chiral system, as one in which there are

two enantiomers that may be interconverted by space inversion, but not by time reversal (Barron, 1986).

Optical rotation changes its sign under parity reversal, but not under time reversal, therefore satisfying Barron's criterion. The electroweak interactions provide an example of a truly chiral influence, and for this reason many authors have restricted their attention to this elementary interaction.

However, under kinetic control irriversibility destroys T-invariance, a situation which allows further possibilities in the context of the life sciences, since life itself is to be understood as a non-equilibrium phenomenon.

It is remarkable that the monomers of proteins and nucleic acids discriminate in favour of one enatiomer. In the case of the amino acids it is the left-handed ones (levo-rotatory, or L-amino acids, that participate in the living cell as monomers of proteins. In the case of nucleic acids it is the dextro-rotatory, or D-ribose that makes up one of the constituents of the sugar-phosphate backbone of he 'double helix'.

George Wald has emphasized (Wald, 1957) that the structure and stability of the polysaccharides, which play a fundamental role in the structure of the plasma membrane demand specific choices amongst the two steroisomers of glucose, which differ in configuration at one asymmetric centre (epimers). Bonner (1991) further points out that the ribose and the 2-deoxyribose monomers of RNA and DNA respectively are exclusively of the D-configuration (this is related to the D-glyceraldehyde). Then it also may be remarked that the glucose monomers of glycogen (a polysaccharide made entirely of glucose units joined together) are exclusively in the D-configuration.

What is more important from the point of view of homochirality is that lipids are no exception in the living world and as in the case of amino acids and nucleotides, lipid chirality is a signature of life.

We wish to underline here that a large class of natural complex lipids, the phosphoglycerides are commonly referred to as *phospholipids*; all the natural 'lecithins' (the old terminology for phosphatidylcholine, one of the most abundant phospholipids in plants and animals) were pointed out by Wald as being dextro-rotatory. At any rate it is clear that the plasma membrane of the Domain Bacteria and Eukarya contain D-glycerol lipids while the Domain Archaea are characterized by membranes containing L-glycerol lipids instead (Kandler, 1995).

2. Evolution at the level of the biomolecules

2.1. A GENERAL VIEW ON ORGANISMS

Having discussed the possible relics of chemical evolution we will dwell upon the forces behind the evolution of the living cell: natural selection, symbiosis, and possibly horizontal gene ransfer. Evolution as a phenomenon of populations is firmly established. The fundamental evolutionary event is a change in the frequency of genes and chromosome configurations in a population, although it is important to underline that the target of selection is the phenotype of a given organism (Mayr, 1983).

We can extrapolate Darwin's well-established concept from the level of populations in two opposite directions. It has been suggested that in this case we may encompass biological evolution (neo-Darwinism) into a whole class of of diverse phenomena which may be referred to as "complex adaptive systems" (Gell-Mann, 1994b), including as well the use of computer software and hardware designed to evolve strategies to make predictions on past observations.

Following this line of thought, the Earth's biota may be included into this larger class of systems, since like other complex-adaptive-system candidates, it may be characterized by the acquisition of information about its environment with which it interacts after processing the information gained; itthe system later acts in the real world according to a well-established model. The ultimate aim of this approach is to lead,

eventually, to further insights gained from phenomena analogous to the living world that may occur in other complex adaptive systems, such as human thought and social evolution. But to date this approach has not helped in biology-related issues.

2.2. EXTRAPOLATION OF EVOLUTION TO MOLECULES

In the concluding paragraph to the introduction of "The Origin of Species", Darwin foresaw that "Natural Selection has been the main but not exclusive means of modification" (Darwin, 1859). For this reason it is particularly fruitful to extrapolate his ideas, not only into more general theoretical approaches, as suggested by the complex-adaptive-systems approach, but also we should entertain extrapolations of Darwin's theories down to the cellular and molecular levels.

Additional factors (in the evolutionary process) have been exposed when a larger repertoire of experimental data is brought into the discussion of cellular and molecular biology. In this manner, at least two other causes of evolution have become evident:

2.2.1. The (Serial) Endosymbiosis Hypothesis. This assumption goes a long way towards establishing the evolution of the eukaryotic cell. Indeed, it is by now almost universally accepted that the mitochondrion and the chloroplast arose as prokaryotic endosymbionts of a simpler eukaryotic cell (Margulis, 1993).Generally, prokaryotes lack nuclear membrane, mitochondrion, chloroplast, and its DNA is normally a single ring-shaped chromosome which is not richly grouped with proteins, although in some cases such as in cyanobacteria there are some histone-like proteins in association with DNA in a nucleoid, and there is at least one example of membrane-bounded nucleoid in the eubacterium *Gemmata obscuriglobus* (Fuerst & Webb, 1991). The compatibility between neo-Darwinism and symbiosis has been discussed in the past (Maynard Smith, 1991).

The key issue is that mutation is not generally adaptive to its causative agent. Hence, adaption arises by natural selection acting on originally non adaptive genetic variation. Symbiosis affords a mechanism by means of which organisms that have evolved separately may come to be subject to natural selection in a single descendant. In this sense symbiosis is a factor not considered by Darwin that must be added to other natural means of transmitting genetic material between organisms, such as sex, and horizontal gene transfer.

2.2.2. Horizontal gene transfer. The flow of genes from parents to offsprings is referred to as vertical gene transfer. There is ample evidence that genes may be transmitted between organisms that are not in the same taxon. Such *horizontal gene transfer* (HGT) may even occur between kingdoms (and even between domains). This may occur in the case of the crown-gall disease; *Agrobacterium tumefaciens*, a prokaryote (Kingdom Monera), does transfer genes to dicots, and even monocots (Kingdom Plantae).

Indeed, the phenomenon occurs so widely (cf., the tables in our recent review Chela-Flores, 1995), that HGT may be an important factor in evolution, although its impact may still be difficult to evaluate Amabile-Cuevas & Chicurel, 1993; Smith *et al.*, 1987).

3. The intimate relation between proteins, nucleic acids and lipids in cell physiology

3.1. FURTHER ASPECTS OF SYMMETRY IN BIOLOGY

It is conceivable that the structure of the chromosome as another form of condensed matter may be subject to a symmetry principle, if the analogy of biological structures with other forms of condensed matter is pursued (Delbruck, 1963; Frohlich, 1977; Chela-Flores, 1985); we shall pursue this point in Sec. 4. Another example may be internal symmetries, a name that is normally reserved by physicists in order to differentiate these kind of symmetries from external space-time symmetries, such as Lorentz invariance.

Internal symmetries may be implemented by the irreducible representations of Lie groups, as in the case of the isotopic spin invariance.

One of the most remote periods that concern the origin of the biomolecules is the one corresponding to the evolution of translation. Nucleic acids, in particular RNA molecules, are capable of serving as the messenger RNA, a first step in the implementation of the transfer of the information from the codons to their correponding amino acids, according to the standard genetic code. More significantly, RNA may also be responsible for the catalysis of other forms of nucleic acids (Cech, 1987; Guerrier-Takeda *et al.*, 1983). RNA itself can also be shown to have other relevant functions. Indeed, further catalytic properties of RNA have been shown in the following experiments:

3.1.1. An RNA-catalyzed hydrolysis of amino acid-RNA bonds has been demonstrated (Picchirilli *et al.*, 1992). Thus a ribozyme-medaited amino-acid charging of an RNA molecule is possible. This is an essential step that occurs in the ribosomes of contemporary cells.

3.1.2. RNA may catalyze the formation of peptide bonds (Noller *et al.*, 1992), thus bringing about the possibility of coevolution of nucleic acids and proteins, as conjectured earlier by Eigen and Schuster.

3.1.3. It has been shown (Mohr *et al.*, 1994) that a protein associated with a *Neurospora* ribozyme can replace an RNA domain in the *Tetrahymena* ribozyme. This step illustrates how the nucleic acids of the RNA world may start evolving a primitive process of translation as the ribozymes may gradually be replaced by proteins.

3.2. THE DEVIATIONS OF THE STANDARD GENETIC CODE

The standard code is known to occur almost universally, but there are variations that were forced upon living organisms such as some eukaryotic organelles which, by symbiosis, may have gven rise to the most trascendental stage in the evolution of life, namely, *eukaryogenesis*, which is the first stage towards the multicelularity of animals and plants.

More importantly, some protozoans and some prokaryotes may deviate from the stndard genetic code (Chela-Flores, 1988):

Indeed, evolutionary changes in termination codon assignments are suggested by the discovery that UAA codes for glutamine in the *Tetrahymena thermophila* gene for the H3 histone (Horowictz & Gorovsky, 1985). This result is at variance with the standard genetic code in which UAA is interpreted as a termination codon; further, the gene for some surface proteins of the ciliate *Paramecium primaurelia* (Caron & Meyer, 1985) has the standard genetic termination codons UAA and UAG coding instead for amino acids.

Besides the above two examples of ciliates of the order Hymenostomata, species of hypotrichous ciliates are known to deviate from the standard genetic code, namely, *Stylonychia lemnae* (Helfteinbein, 1985) and the various species of the ciliate *Euplotes*, such as *E. octocarinatus* (Meyer *et al.*, 1991), *E. crassus* (Harper & Jahn, 1989) and E. *raikovi* (Miceli *et al.*, 1989). In another eukaryote, the yeast *Candida cylindracae* there is a further deviation of the standard code; normally, in the standard code the codon CUG stands for leucine, but the species *C. cylindracae* uses the codon CUG for the amino acid serine (Kawaguchi *et al.*, 1989).

Finally, even amongst the prokaryotes the standard genetic code is not always preserved. In the eubacterium *Escherichia coli* there is a deviation of the standard code: while UGA is used in the standard code as a termination codon, in *E. coli* UGA is used instead to code for selenocysteine (Leinfelder *et al.*, 1988). The Gram-positive bacterium *Mycoplasma* has also been shown to deviate from the standard genetic code. In the case of the species *M. capricolum* the standard-code UGA termination codon is used instead to code for the amino acid tryptophan (Yamao et al., 1985).

The search for clues to the evolution of the genetic codes may be approached by comparison of the usage of anticodons in various organisms and organelles: The GC

content of DNA varies as a result of directional mutation pressure (Jukes *et al.*, 1987). In fact, it is well known that we are now confronted with a variety of genetic codes, each of which may apply to one or many organisms vSome evolutionary arguments may be used in order to find some relationships in this wide variety of codes (Jukes *et al.*, 1987a)

We only wish to emphasize here that there is an ambiguity in the evolution of the genetic codes, which may be illustrated as follows:

3.2.1. According to one viewpoint (Grivell, 1986), divergent genetic codes, as found in ciliated protozoa and *Mycoplasma,* preceded the standard code.

3.2.2. An alternative point of view is that the deviant codes of some protozoans may have formed by the capture of stop codons vand may be of recent origin.

In Table 2 we summarise the main results of the deviations of the Standard Genetic Code.

TABLE 2. Evolutionary changes in termination codons

Organism	*Genus/ species*	*Standard code deviation*
Ciliated protozoan	*Tetrahymena thermophila*	The termination codon UAA codes for glutamine
A ciliate of order Hymenostomata	*Paramecium primaurelia*	UAA and UAG are not termination codons
A hypotrichous ciliate	*Euplotes*	UGA, a termination codon, is translated as cysteine
Eubacterium	*Escherichia coli*	UGA, a termination codon, is translated as selenocysteine
Gram positive bacterium	*Mycoplasma*	UGA termination codon is used to code for tryptophan
Yeast	*Candida cylindracae*	CUG for the amino acid serine instead of leucine

3.3. EVOLUTION OF THE GENETIC CODES

Indeed, it may be possible to approach the intricate question of the evolution of the genetic codes by appealing to the concepts of symmetry. In this context the valuable work of searching for symmetries of the genetic code may be taken as a posssible way of facing the difficulty mentioned above of understanding the antiquity of the genetic codes (Hornos & Hornos, 1993; cf., also comments of Maddox, 1994 and Stewart 1994).In the divergence from the standard genetic code which is known to occur in mitochondria of yeast (Bonitz *et al.*, 1980), *Drosophila* (Brujin, 1983) and human (Barrel *et al.*, 1979), one would question on the basis of the (Serial) Endosymbiosis Hypothesis (Margulis, 1993) that the mitochondria genetic code deviations may prove to be of different symmetry than the standard genetic code.

One possible rationalization of this viewpoint is that the symbiosis may have occurred very early in evolution, the possible scenario being that of a moneran predator invading a larger cell, such as the archaebacterium *Thermoplasma,* thus presumably preceding the appearance of the eukaryotic cells themselves (Margulis & Sagan, 1987), most of which have adopted the standard genetic code. However, there remains the fact that some bacteria and protists do have the standard genetic code; hence, it is not evident how to establish a time chronology in the evolution of the genetic codes of the contemporary living cells.

3.4. THE EARLIEST GENETIC CODE.

More importantly, a similar test may be done with the genetic code in the Phylum/ Division Bryophyta, Class Hepaticae, namely, the chloroplast of the liverwort *Marchantia polymorpha* (Ohayama *et al.*, 1986); (cf., also the discussion of Jukes *et al.*, 1987a). According to the (Serial) Endosymbiosis Hypothesis the origins of some chloroplasts may have been through a complex series of events involving both primary and secondary symbioses, but at least the red algal chloroplast probably derived from a cyanobacterium from by primary symbiosis; there are several reasons for maintaining this hypothesis (Raven, 1970):

Except for their chloroplasts, red algae cells have nothing in common with cyanobacteria organization or biochemistry· besides, chloroplasts of red algae contain a single thylakoid being virtually identical to the cyanobacteria.

As stated in Sec. 1.4 according to the paleontological evidence the earliest date assigned to cellular fossils of the Archean is about 3.47 Gy bp, corresponding to cyanobacteria which may be the oldest organisms on Earth (Schopf, 1993). The question of the antiquity of life on earth has been studied from the point of view of early geochemical evidence, and the possibility of earlier photosynthesizing bacteria may not be ruled out as yet (Schidlowski, 1995).

On the other hand, the question of the age and nature of some of the oldest terrestrial rocks has been considered from the point of view of geochronology (i.e., the application and interpretation of isotopic dating methods in geology). Older records of life are not ruled out, particularly in the rocks that have been studied from the Isua peninsula, some 3.8 Gybp (Moorbath, 1995).We have argued that complete certainty of which organism is a candidate for the most ancient living cell is, at present, a difficult question to answer, although a reasonable guess, based on considerable micropaleontological data, may be that such a candidate is the cyanobacteria.

3.5. COUPLING BETWEEN THE CELL MEMBRANES, DNA AND PROTEINS

To further illustrate the close relation between lipids and the other two biomolecules that are being reviewed in this paper, we may recall that there is a fundamental role for the nuclear envelope in controlling DNA replication within the cell cycle (Blow and Laskey, 1988). In fact using a cell-free replication system from *Xenopus laevis* eggs they have been able to determine which mitotic changes permit DNA to re-replicate. To put this work in perspective we should recall that in eukaryotes the entire genome is replicated precisely once in each cell cycle. It is not possible for DNA to re-replicate until the next cell cycle enters the S-phase after mitosis has been concluded.

This has led to the model in which the control for DNA replication consists of the presence of a cytoplasmic protein ('replicating factor'), which is unable to cross the nuclear membrane (possibly due to its size being bigger than the available nuclear pores). Access to the DNA is gained when the nuclear envelope breaks down at mitosis.

The intimate relationship amongst lipids, DNA and proteins is thus exemplified, as DNA is controlled by the lipid barrier, but the barrier itself is reinforced by a meshwork of filamentous structures in the inner side of the nuclear envelope, constituted mainly by

the lamin B protein. It has been assumed for some time that the nuclear lamina may play a role in gene expression (Newport and Forbes, 1987).

4. Relation in the origin of amino acids, nucleic acids and lipids

4.1. COSMIC ABUNDANCE OF THE 'PRECURSOR BIOMOLECULES'

Given the importance of amphiphilic lipids in the structure of the plasma membrane, we feel that it is appropriate to conclude this lecture with some comments on the origin of the three most important biomolecules.We have to recall that there has been progress in our understanding of the interstellar organic molecules.

There has also been many indications that the origin of life on Earth may not exclude a strong component of extraterrestrial inventories of the precursor molecules that gave rise to the major three biomolecules. About 98% of all matter in the universe is made of hydrogen and helium. The other five biogenic elements C, N, O, S and P make up about 1% of the matter in the universe (cf., Table 3).

TABLE 3: Selected abundances of the elements in the solar photosphere and in carbonaceous (C) chondrites (an abundant type of stony meteorite) analogogue to the prototypical Ivuna (I) meteorite. (Chondrites are normally denoted by a 2-parameter system and (Fegley, 1991).

Atomic number	*Element*	*Abundance in CI chondrites (Si= 10^6 atoms)*	*Abundance in the solar photosphere*
1	Hydrogen (H)	2.79×10^{10}	1×10^{12}
2	Helium (He)	2.72×10^{9}	9.77×10^{10}
6	Carbon (C)	1.01×10^{7}	3.98×10^{8}
7	Nitrogen (N)	3.13×10^{6}	1×10^{8}
8	Oxygen (())	2.38×10^{7}	8.51×10^{8}
14	Silicon (Si)	1×10^{6}	3.55×10^{7}
15	Phosphorous (P)	1.04×10^{4}	2.82×10^{5}
16	Sulfur (S)	5.15×10^{5}	1.62×10^{7}

Some relevant comments on table 3 are needed:

4.1.1. The distribution of oxygen is in contrast to the abundances at hte surface of the Earth, where oxygen is the most abundant element in the crust. Similar problems arise in the terrerstrial planets.

4.1.2. The chemistry of the solar nebula (i.e., the cloud of interstellar gas and dust that condensed to form the Sun and solar system about 5 Gy bp) is essentially the chemistry of the 8 elements H, O, C, N, Mg, Si, Fe, and S, and to a lesser extent Al, Ca, Na and P.

The abundance of biogenic elements would suggest that the major part of the molecules in the universe would be organic. Indeed, out of the hundred molecules that

have been detected either by microwave or infrared spectroscopy 75% are organic (Oro, 1995). Once again, chemical evolution experiments fare well in comparison with the observation from extraterrestrial sources (cf., Table 1 for the earlier illustration). Some of the identified molecular species detected by means of radioastronomy are precisely the same as those shown in the laboratory to be precursor biomolecules.

In Table 4 we illustrate some of these molecules.

TABLE 4: Some interstellar precursor biomolecules (adapted from Oro, 1995)

Interstellar precursor biomolecules	***Formula***
Hydrogen	H_2
Water	H_2O
Ammonia	NH_3
Carbon monoxide	CO
Formaldehyde	CH_2O
Aldehydes	RCHO
Hydrogen sulfide	H_2S
Hydrogen cyanide	HCN
Cyanacetylene	HC_3N
Phosphate	PO_4^{3-}

4.2. FROM THE 'PRECURSOR-BIOMOLECULES' TO THE BIOMOLECULES

Throughout the Mini-symposium we shall encounter many times the major three biomolecules amino acids, nucleic acids and lipids. In table 5 we shall indicate some of the chemical reactions that lead from the precursors biomolecules to the biomolecules.

TABLE 5: Chemical reactions that lead to amino acids, nucleic acids and lipids (adapted from Oro, 1995)

Precursor molecule	*Biomolecule*
Formaldehyde	Ribose, glycerol
$CO + H_2$	Fatty acids
HCN	Purines (adenine, guanine)
H_2NCN (Cyanamide)	Peptides, oligonucleotides and phospholipids
Phosphate	ATP

In all about 12 interstellar molecules have been shown to be precursors of the main biomolecules; table 5 is a brief summary of research that began in the 1950s and extend right to the present. Extensive reviews may be consulted in the Trieste Series on Chemical Evolution (Ponnamperuma and Chela-Flores, 1993; 1995; Chela-Flores *et al.*, 1995; Chela-Flores and Raulin, 1996).

5. Conclusions

We have endeavoured to demonstrate with a variety of illustrations extracted from chemical evolution and molecular biology that the all the biomolecules are intimately related and that the physicist's interest should not be confined exclusively to to proteins We have emphasized particularly that in gene expression the genetic code links proteins and DNA in mutually dependent mechanisms. This symmetry between the structure and functions of proteins and nucleic acids anticipates a deep influence that symmetry has in our understanding of the biomolecules. This is particularly evident when we remark that life is asymmetric in its choice of the constituents of proteins and nucleic acids, but also on the components of lipids and polysaccharides.

These few facts are the backbone of the present Mini-symposium, in which the origins of the asymmetry of the biomolecules and its many implications both to the basic sciences, as well as for its applications, for instance, to pharmacology. We have also attempted to highlight the significant role of that evolution plays in our understanding of the biomolecules. We should appreciate that the origin of the biomolecules depends on evolution at the following levels:

5.1.1. Cosmic,
5.1.2. Chemical, and
5.1.3. Biological (darwinian).

The physical laws that constrain cosmic evolution (the 'Big Bang ' cosmologies) imply a specific choice for the abundance of the chemical elements, predominantly hydrogen and helium. The implication that an initially hot primeval universe is bound to cool down as time increses, in turn implies that galaxies may be formed after the moment of decoupling of radiation and matter; stars then are the source of the heavier elementsa relevant to life as we know it. In the course of this work we have seen, for instance in the chemical analysis of the Murchison Meteorite, that it seems inevitable that cosmic evolution will induce chemical evolution, even prior to the formation of our planetary system. We also indicated that interstellar gas clouds are the cradle of several of the precursors of the biomolecules.

Finally, we have argued that the application of the physical principles to biology is capable of giving us insights to the origin, structure and function of proteins, nucleic acids, lipids and polysaccharides.

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