

## ORIGIN AND EVOLUTION OF ACTUAL BIODIVERSITY

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**Abstract.** Biodiversity is variability of beings in an ecological complex. Since 3.8 billion years ago, there were atmospheric, geological, paleontological and paleoecological “episodes” of crisis and flourishing of different groups of beings. Out of hypothesis and theories on the origin and evolution of life on earth, a special significance have paleontological, systematic, biogeographical, (paleo)ecological, microbiological, botanical and zoological data. Between all prokaryotes, the Archaeobacteria are different by their cell walls and ribosome structure, eukaryotes (protists) resulting from an archaeobacteria and eubacteria symbiosis. The spectacular diversification of protists was possible only after one billion of years of exclusively bacterial beings, after getting the sexual reproduction. Multicellular species resulted from an aggregation between unicellular organisms. Based on analysis and comparison of the gene assemblage of actual beings, molecular phylogeny leads us to a simple reasoning after which, as much as genetic structures of two species are closer, those two species are closer related. The actual biodiversity is a result of remains after mass extinction at the end of Permian and later, at the end of Cretaceous. Today the biodiversity is endangered by a new crisis, increased by the anthropic pressure, as a Biosphere’s “virus”.

**Keywords:** natural history, Archaeobacteria, Eubacteria, prokaryotes, meiosis, diploblastics, triploblastics, biodiversity.

**Rezumat. Originea și evoluția biodiversității actuale.** Biodiversitatea definește variabilitatea viețuitoarelor dintr-un complex ecologic. Începând cu aproximativ 3,8 miliarde de ani au existat “episoade” atmosferice, geologice, paleoclimatice și paleoecologice de criză și de înflorire a diferitelor grupe de viețuitoare. În afara ipotezelor și teoriilor asupra originii și evoluției vieții pe Pământ, o relevanță aparte o au datele paleontologiei, sistematicii, biogeografiei, (paleo)ecologiei, microbiologiei, botanicii și zoologiei. După cercetările genetice se disting etapele precelulară, protocelulară și celulară, dar și o structură ipotetică (progenotul) în care informația ereditară era determinată de acizii ribonucleici. Între procariote, arhebacteriile se deosebesc prin structurile peretelui celular și a ribozomilor, eucariotele (protistele) apărând dintr-o simbioză a unei arhebacterii cu o eubacterie. Diversificarea spectaculoasă a protistelor a fost posibilă abia după un miliard de ani, de viață exclusiv bacteriană, după dobândirea reproducerii sexuate. Speciile pluricelulare au rezultat din agregarea unicelularelor. Pe baza analizei și comparării genelor ansamblului de animale actuale, filogenia moleculară ne conduce spre simplul raționament, după care, cu cât structurile genetice a două specii sunt mai apropiate, cu atât speciile respective trebuie să fie mai strâns înrudite. Biodiversitatea actuală este rezultatul a ceea ce a rămas după extincțiile în masă de la sfârșitul Permianului și apoi de la sfârșitul Cretacicului. Astăzi, biodiversitatea este amenințată de o nouă criză, accentuată de efectele presiunii antropice, ca “virus” al biosferei.

**Cuvinte cheie:** istorie naturală, arhebacterii, eubacterii, procariote, eucariote, meioză, diploblastice, triploblastice, biodiversitate.

### INTRODUCTION

The theme presumes an interrelation of several biological disciplines, with the main available information, at least, on the three large domains of the natural history: Archaea, Bacteria and Eukaryota. Because we cannot survey all the results of each branch of natural history, we will refer briefly to some themes on the origin and evolution of life on Earth, on the one hand, and on the other one, to the creature’s evolution along geological eras up to now, pointing out the entire range of variability of the living organisms.

If talking about the avatars of the biodiversity we have to appeal the results of the deep studies in palaeontology, systematics, ecology, the minute analyses in microbiology, botany, zoology or the related domains (e.g. history of geology), for approaching the proposed theme we are forced to use some classic examples but with “the latest” novelties insertions in our basic knowledge and even to open new directions - themes of imagination stimulation, of thinking and possible application in the next biological research.

In an attempt to gain a global overview of variability among living organisms, from the simplest organic structures to the most evolved plants and animals, we firstly feel to refer to the “Earth biography”, estimated at 4.6-4.8 billion years. Then, micropalaeontology specifies that the first creatures occurred 3.5-3.8 billion years ago, as filamentous bacteria.

If most of the people (specialists or not) accept that the living world has a beginning, the assigning of that beginning is different, according to the philosophical outlook which form the base of the respective explanations. Thus, at first, the **creationist outlook** imposed and still lasts, with a large support, followed by that of the **spontaneous generation** or the **autogenesis theory** (refuted by Pasteur), and then reaching the **evolutionary outlooks**, with several theories and hypotheses.

- **Theory of the biochemical evolution**, represented by OPARIN (1938), at first, and HALDANE (1929, 1932) supports the idea that life is a result of an abiogenic synthesis of the organic substances within special conditions. It might be possible that under those circumstances, several kinds of syntheses “to compete” along the difficult way to the structuring of the protobionts. But, according to those who support this theory, first of all the simple organic compounds synthesized abiogenetically, within the conditions of powerful radiations on Earth, with high temperatures and with a reducing atmosphere.

For the abiogenic formation of the most simple organic substances, the experiment from 1953 made by Miller-Urey (MILLER *et al.*, 1997) became classical, that of submission to the electrical discharge (at 80°C) of the gas mixture, which it is considered to exist in the primary atmosphere: 35% H<sub>2</sub>O + 26% NH<sub>3</sub> + 26% CH<sub>4</sub> + 13% H<sub>2</sub>. After few weeks, on the water film (from the retort – with the imaginary role of the water of the primary ocean) a series of organic substances could be observed: alanine, aspartic acid, glutamic acid, uric, lactic, citric, propanoic acids, etc. FOX & HARADA (1961) synthesized experimentally the uracil. ABELSON (1965) demonstrated that alanine is a stable compound for billions of years, and FOX & DOSE (1972) synthesized amino acids and polypeptides, by heating for 6 - 8 hours at high temperatures (170 °C - 180°C). PONNAMPERUMA & MARINER (1963) obtained ribose and dextro-ribose by the irradiation of the formaldehyde in water, with UV. HARADA K. & FOX (1964) synthesized the amino acids. PONNAMPERUMA (1980) also obtained (besides amino acids) the nitrogenous bases (AGUT) – organic components of RNA, DNA and for ATP; in the presence of H<sub>3</sub>PO<sub>4</sub> and under the action of the UV rays, nitrogenous bases formed mono-, di- adenosine and triphosphate. SCHRAMM (1965) experimentally obtained a series of nucleosides and polynucleotides, and LEHMAN *et al.* (1958) synthesized, in laboratory, the first DNA molecule.

After probation experiments of the abiogenic formation of the macromolecular polymers, the occurrence of the primitive vital structures follows, from the concentration of the macromolecules with vital significance, differently named by different experimenters: coacervates (OPARIN, *op. cit.*); microspheres (FOX *et al.*, *op. cit.*); jeewanu (BAHADUR, 1964), i.e. protocells with properties of metabolism, growing and multiplication; sulfobes, plasmogens, organic microstructures, etc.

The synthesis of such complexes was proved by the Dutch chemist BUNGERBERG de JONG (1932), which experimented colloidal dispersions (phase separation in the solutions of the hydrophilous colloids) and described, for the first time, at the microscope, the structure of Oparin's coacervates, or Fox's microspheres, etc., mentioned above. They are primitive vital structures, individual systems, very instable, but which could become primary living matter, because they have the main features of living; by metabolism, self-reproduction and self-regulation those structures (even if they are instable) opposed entropy, being influenced by natural factors. They were subjected to natural selection and were capable of exchanging matter and energy with external environment.

Thus, the occurrence of the first organisms (protobionts) took place by molecular evolution with a random placement of the monomers, for the purpose of ordering their succession. On the basis of life the nucleic acid molecules or nucleoproteins are present. From the primary cells the prokaryotic cells and then the eukaryote ones occurred.

Commenting the inaccuracy of Oparin's theses, WOESE (1980) appreciated that only the light was the primary source of the energetic processes, and UV radiations, electric discharges and high temperatures functioned as destructive energies for the biological systems. Therefore, life evolution took place on a fundamentally non organic way, in which living systems were sporadically structured, determined by the process which gave them birth. According to WOESE (*op. cit.*) "... the way life occurred has to be similar to the way in which life maintains and evolves".

– **Panspermia theory** was elaborated by ARRHENIUS (1895), considering that life, as matter, is endless. It occurred in those places of the Universe where there were proper conditions. Since Antiquity, ANAXAGORA proposed the term *panspermia*: gr. **pan** – everywhere; **spermio** – germ, and in the 18<sup>th</sup> century, BUFFON (1749 – 1788) talked about the circulation in space of the "life germs". Theory includes two hypotheses:

– of the **cosmosoids** (lithopanspermia) which supports the idea of the transportation of the life germs by meteorites, in which hydrocarbons, aromatic acids, fatty acids, 17 amino-acids, carbohydrates (monoza, glucose), cyclic nitrogen compounds (adenine, guanine). On the origin of these compounds, there are different opinions: some authors support their biogenic origin, and others, on the contrary, support their abiogenic origin.

– **radiopanspermia** (CRICK & ORGEL, 1973), in the universality of the genetic code and of the relatively short period of time between the Earth formation and bacteria occurrence, supports the idea that life occurred on an older planet (with favourable conditions) and the Earth was only "seeded with basic living forms by some intelligent creatures".

– **Absorption theory** sustains the idea that life occurred because of some special minerals (e.g. montmorillonit) with catalytic properties, favouring synthesis reactions, the absorption of the organic compounds, and even their selection in the presence of the unfavourable factors. KATCHASKY & SPITNIK (1947) obtained polymers of the amino-acids, with high molecular masses, suggesting another structuring way of the protobiopolymers.

– **Biostructure theory** (MACOVSKI, 1958), according to which a part of the living protoplasm substances have a special structure, characteristic to the living matter (biostructure); the other part of the protoplasm is the intraplasmatic solution, where biochemical reactions take place. Only in 1981, a team of American specialists observed the microreticulate structure of the cytoplasmic substance.

– "**Cold**" **biosynthesis theory** (SIMIONESCU *et al.*, 1978; MILLER *et al.*, 1997), according to which life would have been occurred on the cold surfaces of the ocean ice or on the ice crystals of the atmosphere; from the basic products (NH<sub>3</sub>, CH<sub>4</sub>, H<sub>2</sub>O) the protobiopolymers synthesize, then the biopolymers, which degrade themselves in monomers and form protocells. Stable microspheres (with  $\varnothing$  of 10 – 50  $\mu$ ) – self-assembled during the thaw, correspond to Fox's microspheres, obtained at high temperatures. In this theory, at least, we see a confirmation of the principle according to which several variant could compete during the abiogenic syntheses of the organic substances.

– **Ribotype theory** (BARBIERI, 1981) supports the idea that life on Earth occurred with the ancestors of the current ribotypes, passing through: **precellular stage** – of ribosome occurrence (from RNA or ribonucleoproteins), of

nucleosides (ribosome coacervate) and of the heterozoids (membrane nucleotides, contaminated with DNA); **protocellular stage** – increasing of the molecular weight of the ribosomes and prokaryotes and microeukaryotes formation; **cellular stage** – formation of archaeobacteria from prokaryotes, and from microeukaryotes – the eukaryotes, with mitochondria and chloroplasts.

– **Progenote theory** (SOGIN *et al.*, 1972; FOX & DOSE, 1972), according to which all current organisms have a monancestral origin (**progenote**), in a hypothetic structure, with a primitive organization, simpler than the current prokaryotic cell. In progenote, hereditary information was determined by the ribonucleic acids.

From progenote, two evolutionary lines started, which determined the **occurrence of the archaeobacteria and eubacteria, to which the evolutionary line of the eukaryotes joined**; each organism keeps itself the historical way of the respective organisms, as informational macromolecules, also named semantides – DNA, RNA and proteins.

There are of course other outlooks, theories and hypotheses on the origin of life on Earth, completing the already known ones, all of them confirming the mentioned principle, of the possible “competition” of several variants of abiogenic syntheses.

In spite of these, although a fully satisfactory theory was not formulated, yet, the problem of life origin does not seem to be very interesting for the beginning of the 21<sup>st</sup> century at the beginning of the 20<sup>th</sup> one. The above-mentioned scenarios seem to be plausible, and the understanding of the respective theories involves a deepening of biochemistry. The degree of this understanding depends on whether we can or cannot accept, respectively, reject the sequence of events, phenomena and facts, which firstly allowed the occurrence and then the evolution of life and creatures along the geological eras.

## DISCUSSIONS

Accepting the explanation of the abiogenic syntheses of the macromolecular polymers and the occurrence of the complexes capable of metabolism, for getting the main features of the living, those pioneers of life should have found the modality of getting energy and reproducing themselves. Or, when referring to billions of years ago, when the Earth's atmosphere was devoid of oxygen, but the sun provided a lot of energy and the oceans provided energy from sulphides, we can say that procuring power and growth was even possible. On the other hand, rocky surfaces were covered with metabolising films, which could grow but they could not reproduce. So, a great difficulty of the basic organic structures was the invention of the genetic information transmission from one generation to another.

For the time being, it is known that DNA has enzymatic qualities and therefore it could be selected for the characteristic. But since his role in reproduction is secondary, a living world based on RNA may have been before that based on DNA, when a protein synthesis already existed; but the efficiency of the protein synthesis, made with the DNA, was absent.

But since life arose on Earth once conditions became suitable (about 3.8 billions of years ago), it seems the appearance of life was not too difficult, within the temperature and chemistry conditions of the environment of that time. As yet, we do not have fossils from the period 3.8 – 3.5 billions years ago, but fossil rocks of 3.5 years old includes a bacterial biota, remarkable rich.

Over the next billion of years, prokaryotes were the single form of life, otherwise stunningly diverse: cyanobacteria, gram-negative bacteria, gram-positive bacteria, purple bacteria, archaeobacteria. There are still questions on their relationship, because, bacteria do not have neither biological species nor sexual reproduction.

On the other hand, bacteria exchange genes between them, sometimes even entire blocks of genes. By the *lateral transfer*, a bacterium which belongs to a subdivision (e.g. of the gram-negative ones) may have a set of genes specific to a completely different subdivision.

Therefore, in the case of bacteria it is difficult, even impossible, to build hierarchically ordered trees, as in the case of the eukaryotes. WOESE & GUPTA (1981) discussed the place of the Archaeobacteria in the long process of appearance of the cellular structures and WOESE *et al.* (1980) made a phylogenetic analysis of microplasma.

On the other hand, the specialists with different opinion agree with two taxonomical philosophies: classification of the prokaryotes according to their differentiation degree; - taxa are spread according to the succession of the branching points of the phylogenetic tree, after Hennig's ordering system.

This contradiction regards especially the archaeobacteria position, which distinguishes from other bacteria by the structure of the cellular wall and ribosomes structure, although they are typical prokaryotes.

CAVALIER-SMITH *et al.* (1998) classified the Archaeobacteria as one of the four great subdivisions of bacteria. Archaeobacteria do not distinguish from other types of bacteria more than protists distinguish one another, or more precisely, they have the ribosome structure as the eukaryotes. **The first eukaryotes appeared from a symbiosis of an archaeobacteria with an eubacteria** and then by the formation of a chimera of the two symbionts. This explains why the new taxon (**Eukaryota**) combines both the characteristics of the Archaeobacteria and those of the Eubacteria.

It cannot be specified which bacteria participated to this process. Spirochetes must have been engaged to provide cilia. MARGULIS (1970 and 1996) asserted that in a protest five different bacterial genomes can be recognized.

It is certain that the first chimera got additional genomes, by unilateral transfer of genes. The frequency of this transfer, including among prokaryotes so far as Eubacteria and Archaeobacteria, represents the major difficulty in the reconstruction of the prokaryote phylogeny.

In the biodiversity saga, it can be said that **the most important and dramatic event from the entire history of life on Earth was the eukaryotes occurrence**, after a billion of years of exclusively bacterial life. From that time on the appearance of all more complex organisms was possible: protists, fungi, algae, plants, animals. The achievements of the descendants of the first eukaryotes consist in: - the structure of the nucleated cells; - sexual reproduction; - meiosis.

Because of their way of life (on organic detritus or as parasites), prokaryotes remained (after eukaryotes appearance) extremely spread and they became even more numerous. A series of calculations was made according to which the biomass of all prokaryotes on Earth is as high as that of the eukaryotes!

Bacteria distinguish from the eukaryotes by some common characteristics: - they do not have nucleus; - DNA is placed in gonophores; - chromosomes do not contain proteins; - they do not reproduce sexually; - cellular division is achieved by simple fission or by budding (without mitosis or meiosis); - bacterial flagella are formed of flagellin protein and they are of a rotary type; - cells are usually small (1 – 10  $\mu$ ), some of them creating some colonial aggregates (mitochondria, chloroplasts, etc.).

The rich world of prokaryotes is difficult to be grouped. One of the subdivisions (Archaeobacteria) consists of genera adapted to the extreme conditions of environment (e.g. in great oceanic depths, near thermal springs with sulphur, in extremely salty environments, etc.), but also of genera which are distributed everywhere, including in the sea and ocean water. BRADLEY (2009), commenting the analysis of the recent discoveries in the vicinity of the oceanic ridges (where water can reach 400°C, and with an acid pH (“as the lemon juice”), with rich precipitations of sulphates, iron, copper and zinc, at only 15 km far from those springs water reaches 90°C, pH becomes alkaline and there is a fauna (e.g. vestimentiferan *Riftia*) which lives in symbiosis with a series of internal bacteria, capable to metabolize H<sub>2</sub>S – otherwise a toxic gas. The discovery of those forms of life, independent of the sun power, raised the question whether the origin of life on Earth might be even from such chemical conditions.

Knowing that the first fossil prokaryotes (3.5 billion years old) were cyanobacteria, we can say that they have a morphological stasis; about 1/3 of the fossil primitive species of prokaryotes do not differentiate morphologically from the current ones, and almost all fossil cyanobacteria can be classified in modern genera.

Long preservation of the morphologic homogeneity is attributed to the asexual reproduction, although cyanobacteria have huge populations, capable to live in highly variable environmental conditions, even in extreme environments.

Eukaryotes differ from prokaryotes in that they possess a nucleus surrounded by membrane which contains individual chromosomes. I mentioned that the first eukaryote formed due to a symbiosis (chimera) of an archaeobacterium with a eubacterium. This way of occurrence was deduced from the partial archaeobacterial composition and partially eubacterial of the eukaryotic genome (MARGULIS *et al.*, 2000).

The new eukaryotic cell acquired later different symbionts, as cellular organelles (mitochondria and chloroplasts – in plants). Organelles were acquired successively because some primitive eukaryotes (but which are still in a living state) are devoid of mitochondria or other intracellular organelles. It is not known precisely how nucleus appeared, in which the chromosomes are placed inside a membrane; probably, when the nucleus formed, the symbiosis was not implied.

Mitochondria derived from the alpha subdivision of the purple bacteria (proteobacteria), and the chloroplasts of plants – from cyanobacteria.

**Protists.** The primitive unicellular eukaryotes (with or without cellular organelles) are known collectively as protists, included in different kingdoms: protozoans, sulphonamides, the simplest representatives of plants, algae, fungi and animals. Most of the protists do not have cellular organelles because they lost them along their evolution. Eukaryotes have diversified spectacularly as they occur, about 2.7 billions of years ago. MARGULIS & SCHWARTZ (1998) recognized 36 protist phyla: amoebae, microsporidians, mucilaginous molds, dinoflagellates, ciliates, sporozoans, cryptomonads, flagellates, xanthophytes, diatoms, brown algae (some of them pluricellular), oomycetes, myxosporea, red algae, green algae, radiolarians and other 20 phyla, less known.

But the incomplete understanding of the phylogenetic relationships among the unicellular eukaryotes is demonstrated also by another modern classification, which divides the protists in 80 phyla. There is the risk that Protista taxa not to be recognized because of its extreme heterogeneity; we are far from a stable classification of protists, which will impose the methods of the molecular biology.

The most primitive fossil unicellular eukaryotes (protists and algae) dates from 1.7 billions of years, but diverse investigation methods allow us to presume that they occurred about 1 billion years earlier. The diversity of the primitive eukaryotes remained limited for 800 million years (in the period 1700 million years and 900 million years). Then, it has grown rapidly because in Cambrian a real **explosion of protists** took place, this time proved by the presence of numerous microfossils.

**Multicellularity** occurred repeatedly during evolution. Among bacteria there are numerous precursors of the multicellular organisms. The first step to multicellularity was the growth in size of the aggregates. This was pointed out in several tens of groups of unicellular protists, algae and fungi, finally leading to a labour division between the cells of such aggregates, which then associated in an authentic multicellular organism.

Primitive eukaryotes were unicellular, reason why they were defined as unicellular eukaryotes, for a long period of time. Then it was discovered that both the unicellular plants (e.g. green algae) and unicellular animals

(protozoans), and unicellular fungi on the one hand, and some taxa with unicellular species, on the other one (e.g. brown algae – **Phaeophyta**, and red algae – **Rhodophyta**) also include multicellular species, a classical example being the Giant kelp (*Macrocystis pyrifera*) from protists (brown alga from the Californian coast) which begins its life cycle as a microscopic spore and grows up to 60 - 100 m long.

In conclusion, a series of multicellular forms are widely distributed among the fundamentally unicellular taxa. Sometimes, even bacteria aggregate in large masses of cells. Multicellularity reached its peak in three large phyla: plants (metaphytes), fungi and animals (metazoans). Only 40 years ago, in the biological classifications, fungi and algae were among plants, and protozoans were included in the animal kingdom; today, unicellular organisms belong to protists.

I mentioned above that **in the Cambrian there was an explosion of the animal life**. Indeed, 542 million years ago there was a series of digging, swimming, creeping, fixed animals, some of them being predators, others were hunted. In fact, on the marine bottom of that time, the main modern ecosystems were developing. BRASIER (1992) referred to the basis of the Cambrian “explosion”.

ADAM SEDGWICK (president of the Geological Society of London) and his colleague, RODERICK MURCHINSON, studied (in the 19<sup>th</sup> century) rocky outcrops of Wales. Digging the upper strata, MURCHINSON named them Silurian, while SEDGWICK, studying the lower ones, named them Cambrian (lat. Cambria = Wales). Besides the popular conferences of the geological discoveries, both specialists published aspects on the Silurian and Cambrian strata succession from England and Wales, in 1835.

The rocks of Cambrian age have been dated at 542 – 488 million years old and are rich in much diversified animal fossils. This was surprising because older strata lack of such richness. SEDGWICK’s young student of that time, CHARLES DARWIN, looked for an explanation of these differences, and in 1859 (in **Origin of species**), he wrote: “**Why we do not find remains of the fossil ancestors from Cambrian under these geological strata?**”

Later, geologists found the fossils of some multicellular organisms in geological strata older than Cambrian. Such deposits were discovered in Australia, Canada, Namibia, Russia, 575 – 542 million years old and with a fauna formed of organisms with soft bodies, plumose or disc-shaped, which could not be classified in any of the current phyla. Palaeontologists named them **fauna of Ediacara**, after the name of the Australian in which it was discovered. Another deposit (Doushantuo) from China has an older fauna (600 – 570 million years), with sponges and cnidarians preserved in good conditions.

But the Cambrian did not remain a unique period for the diversification of the animal world. In the Canadian site Burgess (dated of 505 million of years) and in the site Chengjiang from southern China (dated 520 million years) it was discovered that along some tens of millions of years marine world enriched with new forms of life which gave rise to most of the current fauna; about 40% of the current phyla exist since the beginning of the Cambrian. It is about arthropods, molluscs, echinoderms and chordate, as ancestors of the vertebrates. Although sponges lack the body symmetry, and cnidarians have radial symmetry, the other animals are characterized by bilateral symmetry.

Some of such Cambrian animals (with bilateral symmetry), enigmatic at first sight, probably belong to the evolutionary lines in which the current phyla originate. The identification among them of some predators demonstrates the presence of some complex trophic relationships since then. In this regard, in site Chengjiang 180 species were discovered, 40% of them being arthropods, still occurred in marine ecosystems. Their morphology indicates a high motive and sense capacity (especially the visual one). Out of them, about 20% do not have current representatives: sponges (13%), priapulid worms (8%), then brachiopods, molluscs, chordates – ancestors of the vertebrates.

VANNIER (2009) demonstrated that there were over 90 species of that time at the water-sediment limit, this representing a fundamental difference from current marine world. Some organisms (sponges, ascidia) were fixed and filtered water for feeding with particles in suspension. Others (bradoriides, small bivalve arthropods of only some mm) were on the sea bottom and recycled organic matter. Molluscs “grazed” the bacteria blanket, and arthropods hunted; the presence of such kind of predators, suggested by the structure of the prehensile appendages of numerous species, is confirmed by the presence of the microscopic trilobites from the digestive tract of some of them. Preserved traces and galleries on the marine bottom show that some animals lived in the sediment, as the priapulid worms, discovered in the Chengjiang deposit. This possibility was verified and confirmed in laboratory experiments, but also by the comparison of the Cambrian traces with those of the current priapulid worms from the bottoms devoid of oxygen of the Swedish fiords. Morphology of the fossils also shows that the Cambrian priapulid worms were predators, which probably fed on bradoriides and hyolithes – animals resembling small molluscs, conic-shaped. Colonization of the water blanket by the medusoid organisms was made by organisms which resemble the current ones. The great predator *Anomalocaris* was 1 meter long and its morphology placed it among the arthropod swimmers of the neighbourhood of the sea bottom.

The question is if there was zooplankton in the Cambrian oceans – those inferior animals incapable to oppose to the spreading effect of the currents, and which, in current oceans, it has an essential part in trophic relationships. At Chengjiang, fossils of worm-like animals were discovered (chaetognaths), whose current representatives belong to the carnivorous zooplankton – predators of herbivorous zooplankton. Also, other Cambrian deposits have animal fossils which had oral crochets. Chaetognaths could be a key element in the first trophic relationships of the sea. It might be possible that the Cambrian animals not to colonize marine domain, they concentrating to the interface water-sediment or only to the lower level of the water column.

So, at the beginning of Cambrian, predator animals were in sediments, on the marine bottom and above it, only in some few meters of the water column. That means that the non-predator animals were strongly subjected to a selective pressure. The interaction development between prey and predator could favour **that diversification, mentioned for Cambrian, preys evolving under the pressure of predators, and predators evolving according to the available resources**. Interactions between prey and predator favoured the diversification of the creatures.

Nevertheless, the release of this diversification remains rather mysterious, because it is not known exactly where the animals came from. Fossil remains of their ancestors were not found. For instance, the strange fossil *Vernanimalcula guizhouena* of only 200  $\mu$  in size and with bilateral symmetry, discovered in the Doushantuo deposit, is questionable regarding its animal nature. The link between the first animal communities from Ediacarian and those from Cambrian is better known today.

ERWIN (2009) asked whether Cambrian explosion was preceded by another explosion of biodiversity (e.g. in the Precambrian). This hypothesis was released by a team of American palaeontologists, in January 2008, on the occasion of the investigation of the Ediacarian fauna, occurred 575 million years ago. SHEN, DONG, XIAO and KOWALEWSKI (2008), and other scientists of the Polytechnic Institute of Virginia, studied over 270 species originating in 30 sites from all continents. These species were divided into three groups, according to the age of the rocks where they were found: - "Avalon", for those dated between 575 – 560 million years; - "White Sea", for those of 560 – 550 million of years; - "Nama", for the newest ones, estimated at 550 – 542 million years.

At the first sight, this fauna seems to be little diversified. But the American palaeontologists compared 50 morphologic criteria and pointed out statistically the numerous anatomic disappearing from the first group (Avalon). They were the witnesses of a major evolutionary event, occurred at the beginning of the Ediacarian and which would have generated the diversification of living animals of that time. We shall see whether this diversification can be compared with the Cambrian explosion of biodiversity. But what is precisely known is that the diversity of the creature forms corresponds to the diversity of the organization forms.

In the same biodiversity epic there is another episode which refers to the Earth's major environmental upsetting. Between 635 – 530 million years ago, a huge glaciation installed, which covered almost the entire planet. Surviving species evolved in the oceans which gradually oxygenated. In addition, from the erosion of the new mountain chains, those species took advantage from a great supply of nutrients. We may say that the Earth was in the middle of some drastic transformations and that the diversity of the creatures occurred in the newly oxygenated oceans together with the installation of large scale glaciations.

It is obvious that there were not good life conditions in the shallow cold oceans, rich in sulphur and poor in oxygen. The ground emerged from water (Pangea) and divided into two blocks (Laurasia and Gondwana). 120 million years ago, Gondwana underwent an intense greenhouse effect, and the ocean oxygenated in depth. This occurred when the aquatic fauna opened new anatomical diagrams, as morphologic innovations.

But which is the link between the environmental changes and morphological innovations?

Geologists' stake is to distinguish different environmental conditions in order to understand what could generate or, at least, favour the evolution of the animal world. In other words, it had to be distinguished the causal connections of the temporal correlations.

Studies on fossils, as well as those on the genetic evolution of current genera and species show that cnidarian diversification (corrals, jelly fish, etc.) and of the Ediacarian organisms began 575 million years ago, while in Cambrian (we mentioned above) the ecosystems were organizing. At least 20 million years later, bilateral organisms began to diversify. At the same time, numerous independent branches were deeply affected, and this demonstrates that the releaser was an event of a planetary scale.

With the acquisition of those new morphological features, the organization of creatures was totally upset. It was very particular in the history of Earth, because surely it was born also within particular conditions. In 1998, PAUL HOFFMAN, that time working at the Harward University, was the first who suggested that 650 million years ago, our planet new an environment with extreme life conditions, which determined the subsequent diversification of the biodiversity. In fact, it is about that ice age, named **marinoean**, of an uncommon scale and lasted 15 million years; under these circumstances only a few small areas on the Equator line remained unfrozen. Viewed from space, the Earth looked entirely white. Biological production was possible only in void that time. HALVERSON *et al.* (2002), HOFFMAN *et al.* (2007), SCHRAG (2007), SCHRAG *et al.* (2002), also from Harward, imagined that Earth looked like a snowball that time. Glaciation might have begun 1 billion years before (at the end of Neoproterozoic) and would have ended at the beginning of Cambrian, i.e. 542 million years ago. After the appearance of rocks, remarked by geologists in Namibia, marinoean glaciation should have been global. The rocks with CaCO<sub>3</sub> (chalk) and with magnesium (dolomites) were covered by the glacial deposits. Such carbonates, called "coverage", are very rare. Their origin was understood only at the end of 1990, when several geologists analysed the magnetic "signature" of the glacial deposits. These deposits were formed near the equator more than near the poles. "Coverage" carbonates belong to the marine sediments. So, the glaciation was formed at the sea level. But how could "coverage" carbonates formed near the Equator? SCHRAG (op. cit.), HOFFMAN *et al.* (op. cit.) inspired after the proposal by the Americans KOPP, KIRSHVINK, HILBURN and NASH (2005) few years ago and they reached the conclusion that the Earth might have been as a snowball, polar ice caps extending almost on the entire planet. That ice age could finish because of a huge greenhouse effect, generated by the CO<sub>2</sub> emitted from volcanic eruptions.

Dating precisely the volcanic ash layer discovered in the glacial rocks from China, Namibia and Oman, HOFFMAN *et al.* (2007), then BOWRING *et al.* (2007) from the Institute of Technology of Massachusetts, found a full concordance between spatial and temporal data. In both places, ashes were deposited 635 million years ago.

Also in both places, thawing was simultaneous (global), especially that they were less remote than they are today.

Although according to ERWIN (2009) the third scenario of the “snowball” is still controversial, it is the single one which can explain the formation of the “coverage” carbonates at Equator.

Nobody doubts that the glaciations influenced the living world. That time there were only some ecologic niches, which allowed the evolution of the animal world. Glaciation and then quickly thaw could generate the disappearance of several species and the adaptation of a part of the populations which resisted, within the conditions in which the evolution speed increased very much. Also there is a full concordance between glaciation data and explosive emergence of new species. For example, the traces of the first sponges are 635 million years old – at the end of marinoean glaciation. The first Ediacarian organisms occurred 575 million years ago – at the end of a subsequent glaciation (Gaskier), and doubtless, less severe. All these are only correlations and not necessarily causal relationships. Over all, though the hypothesis of the “Snowball” Earth is seductive, it cannot explain the formidable combination of morphological innovations, observed in Ediacarian and Cambrian organisms.

But outside the temporal correlation between glaciation and diversification of living creatures, other courses have to be explored. For instance, the physical-chemical characteristic of the oceans and atmosphere seems to be more promising. Using the technique of isotopic chemistry, the geo-chemists can understand the state of the oceans and of the atmosphere of that time. In fact, **the rocks from the end of the Neoproterozoic registered chemical changes of the environment inside them.** Using a mass spectrometer, geologists have measured the content of different forms (isotopes) of carbon and sulphur and described the evolution of the cycles of these elements. Also, different geological and biological processes that best fit the discovered isotopic composition can be distinguished. The analysis of the rock content in carbon allows the study of the evolution of the organic reservoir of carbon, which gathers all living creatures and the carbon which derived, as coal. This reservoir is different from limestone and other inorganic forms of carbon by the content in Carbon-12 and Carbon-13. In inorganic reservoirs, content ratio of these two isotopes of carbon remains constant. On the contrary, photosynthesis enriches organic reservoir, with Carbon-12. An increase in the load of Carbon-12 in rocks implies an increase of the organic reservoir, because of the photosynthesis activity of the marine phytoplankton. But, by this process, the oxygen content of the ocean increased.

Regarding the sulphur, although the explanation is more complicated, the ratios between its isotopes allowed the increase of the sulphates quantity in the ocean, quantity also linked by the content of oxygen from the atmosphere.

The increase of the oxygen content of oceans and atmosphere – till then a virtual gas for living creatures – represents the key of the animal diversification enigma. Ediacarian and the beginning of Cambrian were eras in which the changes of the carbon and sulphur cycles were the most important from the entire history of the Earth. Sulphur isotopes indicate significant changes immediately after marinoean glaciations, about 635 million years ago. The rocks from Oman, Namibia and China indicate the same important changes 560 million years ago, when it took place the most important increase of Carbon-12, of the Earth’s history; indirectly, an increase of the oxygen content of the oceans took place, too. It is generally accepted that regular increase oxygen levels in the ocean imposed 600 million years ago.

But, which was the origin of CO<sub>2</sub> release? With no consensus on the natural phenomena of the past, a question rises – where the respective growths of CO<sub>2</sub> came from? Some people attribute to massive discharges of CO<sub>2</sub> in the atmosphere, during the glaciations. When the glacial caps melted, marine phytoplankton absorbed CO<sub>2</sub> and developed. Thus, carbon was deposited progressively in the marine organic matter, which led to the O<sub>2</sub> discharge in the oceans. Australian biochemists GRAHAM LOGAN *et al.* proposed an explanation in 1995, according to which the animals which acquired the digestive tract in their structure and were far from the coast, swallowed the organic matter from shallow waters and eliminated faeces rich in oxygen. But because of their weight, they were trained in deep oceans. From this moment on, the limit between the waters rich in oxygen and those rich in sulphur changed, passing in the shallow waters of the sediments. Thus, the oceans were more oxygenated. Even if the idea is seductive, it seems that the oxygen content had begun to increase before the animals with digestive tract occurred. Therefore the process described by GRAHAM LOGAN *et al.* (op. cit.) is not the cause of the discharge increase or in any case they were less amplified.

In 2007, ERIK SPERLING and D. PISANI from Yale University and KEVIN PETERSON from Dartmouth College of New Hampshire pointed out the importance of sponges – big sacs with collagen, which could transport large quantities of carbon. Colonizing the waters deeper and deeper, sponges transported progressively the oxygen in oceans. But it is not known if this method was sufficient for ventilation.

Another way would have been that after glaciations the species occurred only from restrained ecological niches. In order to develop and to evolve, they needed an important quantity of nutrients, mostly generated by the tectonic of the continental plates. 600 million years ago, the continents which had begun to split apart by the fragmentation of the supercontinent Rodinia, 150 million years ago, this time they group again, the two blocks merged to form Gondwana. By that contact, the African plates merged and formed Pan African chain. The consequence of that orogenesis was increased erosion. At the same time, three large islands separated: Laurentia, Northern Europe and Siberia. Those events, correlated with the marinoean deglaciation because of the great quantity of CO<sub>2</sub>, also increased the erosion process, which was confirmed by the ratio between the strontium-87 and strontium-86 content – a ratio which depends on the erosion degree and the material emergence of the crust mantle into the ocean ridges. 580 million

years ago (at the end of Gaskier glaciations), the erosion increase was really strong. Erosion process increased continuously till the beginning of the Cambrian, large amounts of iron, calcium, phosphorus and bicarbonates being released. These nutrients have contributed to increased phytoplankton production in reheated and freshly oxygenated ocean, finally helping the evolution of the first animals.

Global glaciations, followed by a strong greenhouse effect, an increase of the oxygen in oceans and high availability of nutrients could help together the diversification of the animals known on the Earth, between 600 – 530 million years ago. At the same time, each environmental change taken separately was not sufficient. Therefore, a conjugation of all those events was necessary – a unique conjugation in the Earth's history, which was the basis of the diversification of creatures.

Besides the two important biodiversity developments from Precambrian and Cambrian, there was a third explosion, also in the Cambrian – probably the most interesting from the biodiversity epic, in terms of further development of creatures over geological eras.

For the time being, biologists do not have any fossil which might represent the last common ancestor of animals with bilateral symmetry, whose diversity “exploded” in the Cambrian and which could be the origin of most of the current animals. To achieve a robot-portrait we appeal to molecular biology and development genetics.

A question occurs, if we can find out to whom the last modern animal resembles?

The last common ancestor of most of the current animals and humans occurred in the Precambrian – over 542 million years ago. According to present hypotheses, it would have resembled either a platyhelminth or an annelid.

*Convolutriloba longifissura* was a wide oval worm, of a living colour, which lived in marine shallow waters, with a sandy bottom. In its movements he seemed floating in water because of the flaps of the numerous small cilia, which covered entirely its epidermis. That graceful animal belonged to acoelomates group, which had a characteristic simple anatomy, today being represented by more than 100 species. Their flattened body is clearly structured by a front and a rear part, without a distinguished head; these animals do not have a mouth, their digestive system opening either frontally or ventrally, or in the rear extremity of the body. Also, acoelomates are devoid of digestive tract, have only a compact cellular mass, important for digestion, which fills a large part of the body and communicates with the exterior by mouth – which can be also the anus. Their nervous system is diffuse, located in the epidermis, without brain and nerve trunk.

For BALAVOINE (2009), *Convolutriloba* and its congeners could solve, at least partially, an enigma of the evolution, that is – to know who the last common ancestor of the animals with bilateral symmetry resembles. Diversity of these animals “exploded” in the Cambrian and from them all current animals descended, except sponges and cnidarians. This assertion has been supported recently by MARK MARTINDALE and ANDREAS HEJNOL (2009) - from the University of Hawaii.

But, other scientists have developed a second hypothesis, according to which the ancestor of all animals with bilateral symmetry had a series of common features with the more complex structure animals than the acoelomates; besides, annelids are devoid of a condensed nervous system and a complete digestive tract, with distinct oral and anal openings. Therefore, there is a concentration of the efforts in order to decide if the ancestors of the bilateral animals was a platyhelminth or an annelid.

The understanding of the evolutionary history of animals with bilateral symmetry and tracing of the evolution line is a more difficult task because the Precambrian ancestors of the current animals vanished long time ago. All science can get are the clues on the form of these ancestors. It is not about paleontological clues. Fossil archives demonstrate the abundance of the organisms with bilateral symmetry in the Cambrian, 540 million years ago. But, this abundance proves that first of them occurred before, maybe even in the Precambrian. For the time being, we do not have any indisputable fossil of that age. It should be found other means for making a robot-portrait of the ancestor of the bilateral animals: molecular phylogeny and development genetics.

Molecular phylogeny presumes the analysis and comparison of the genes of the current animal assembly, which allow elucidation of kinship ties between the major lines of animals, following a simple reason: the more genetic structures of two species are closer the more the species have to be stronger related. What the molecular phylogenies tell us today regarding the large groups of animals is plausible, reliable, and normal. At the basis of the phylogenetic tree are sponges, which have neither real tissues nor symmetric plan. Then, cnidarians are formed of distinct tissues, but devoid of complex organs, but have structures displayed after a radial symmetry.

Then the separation of animals with bilateral symmetry (**Bilateralialia**), which are related and divide in two large groups followed: 1. protostomes with a) - ecdysozoans (arthropods and other animals with cuticle, with a body which grows by subsequent shedding); b) - trochozoans (molluscs and especially annelid worms); 2. deuterostomes, in which echinoderms, hemichordates, urochordates and chordates.

Some phyla remain, being hard to be included in any classification. Acoelomates are a good example. Their first phylogenetic analyses were made in 1999, by JAUME BAGUÑA from the University of Barcelona, together with his Spanish and English colleagues. This analysis was made on a single and unique gene – codon of an RNA incorporating ribosomes – RNA 18S. Two conclusions were reached: - that acoelomates do not belong to the Platyhelminthes Phylum, in which they were included till recently only due to their morphological features; - that the acoelomates would be the most related creatures to the first organisms with bilateral symmetry. That means that their evolutionary line emerged from the phylogenetic tree, between cnidarians and the animals with bilateral symmetry.



Specifying the anagenesis and cladogenesis are mostly independent processes, MAYR (2001) pointed out DARWIN's observation (1859), according to which in the evolution of living creatures there are two aspects – one of modification of the phyletic line from an ancestral state to another state, a derived one (anagenesis), and the other aspect consisting in the occurrence of a new branch (clade) of the phylogenetic tree (cladogenesis) – a process which gives birth to the biodiversity by speciation and removal by the ancestral type. According to SCHACHERER (2009), by the sequencing techniques of the genome, the molecular biology has deciphered the role of genes and especially the role of their duplication, as a function of “engine” of evolution. If genome evolves by the acquisition of new genes, this acquisition is achieved mostly by the duplication of pre-existing genes or groups of genes.

MARK MARTINDALE and ANDREAS HEJNOL (1999) brought other arguments, i. e. acoelomates have features which are found not only in sponges and cnidarians (from the basis of the phylogenetic tree for the animal kingdom) but also in several groups of animals with bilateral symmetry. For instance, the absence of the anus in sponges, cnidarians, acoelomates and plathelminthes. For Martindale and Hejnol (op. cit.), who considered that the acoelomates evolved a little along the 500 million years, and their common features would be plesiomorphs, morphology of the current acoelomates would reflect the morphology of the last common ancestor of the animals with bilateral symmetry. More than that, by the term *Urbilateria* it is designated the hypothetical ancestor of all animals which have bilateral symmetry with respect to the central axis of their body (from worms to mammals), supposed to be acquired over 600 million years ago (Fig. 1).

Preoccupied to understand the phenomenon by which a primitive organism of an acoelomate type would have evolved in more complex organisms, the two biologists appealed to the development genetics. They investigated if in *Convolutriloba longifissura* there are genes which lead to the formation of the organs in animals with bilateral symmetry. First of all, a question occurred: if these organs exist in *Convolutriloba* and then, if they exist, which is their part?

MARK MARTINDALE and ANDREAS HEJNOL (2009) demonstrated that these genes are real, and are always present in acoelomates, but express themselves in a less specific way than in bilateral animals. An example refers to the gene *Emx*, specifically expressed in the anterior part of the nervous system in bilateral animals; in acoelomates it is expressed along the embryo. The two biologists reached the conclusion that the **ancestor of the animals with bilateral symmetry had “generalist” genes** which later specialized in the development of one or another of the organs of bilateral symmetry animals.

These genetic data are interesting and simultaneous in correlations, but the interpretation given by MARK MARTINDALE and ANDREAS HEJNOL (2009) is disputable. The strongest counterargument was brought by the new phylogenetic studies, recently made by HERVÉ *et al.* (2007), from the University of Montreal, among his collaborators also being JAUME BAGUÑA, who wrote about bilateral symmetry of the first organisms with another team of specialists, in 1999.

PHILIPPE *et al.* (2007) concentrated on 68 protein coding genes from an acoelomate and on other 51 animal species belonging to 15 different phyla, instead to refer to the coding gene ARN 18S. Starting from their analysis, published in August 2007, it resulted that the acoelomates are not at the base of the animals with bilateral symmetry! The lack of knowledge where they can be placed in the phylogenetic tree is explained by the fact that these genes do not evolve in the same rhythm, in all organisms. „Molecular chronometer” rotates faster or slower from a phylum to another and indicates that in the case of acoelomates any record was broken: their genes evolve so rapidly then it is very difficult to place these animals in the phylogenetic tree and they remain *incertae sedis*. With their evolution speed, also very rapid, it is improbable that acoelomates to be the descendants of “the missing link” between cnidarians and bilateral symmetry animals. But how can we determine our distant ancestor portrait?

In 1980 and 2006, DETLEV ARENDT from the Laboratory of Molecular Biology from Heidelberg and then ARENDT & NUBLER-JUNG (1994), dealing with the **development genetics**, discovered that in the developing of the current animals several genes are involved, which are present in very different phyla and which often plays a surprisingly comparable role. A founding example is that of the *Hox* architect genes. They are grouped on the same chromosome, very close one another, in a very precise order: each of them controls the morphology of a specific part of the body, along the anterior-posterior axis of the animal, in an order similar to the order of the genes on the chromosome. Thus, the insects have a complex in which each *Hox* gene controls the precise form of one or several segments, while in tetrapod vertebrates there are four complexes of *Hox* genes which, among others, control the form of vertebrae and ribs. It was discovered that each *Hox* complex in vertebrates is arranged like a *Hox* complex in insects.

But, from our point of view, this assemblage of genes is very complicated for occurring twice, independent in evolution. Their presence in vertebrates and in insects suggests that such a complex of *Hox* genes already existed in the last common ancestor of these animals. In other words, it was present in the ancestors of the animals with bilateral symmetry. Also, it suggests that the ancestor already had a differentiated axis. KULAKOVA *et al.* (2006) pointed out the importance of *Hox* genes in larval development of the polychaetes *Nereis virens* and *Platynereis dumerillii*.

Today we know many other examples of similarities, at a large scale of the embryonic functions of genes. The most recent example refers to the nervous system. For 200 years biologists have been discussing the theme: why in the arthropod, annelid and mollusc world the nervous system is placed ventrally and why in vertebrates, dorsally? Especially, because in both cases, it is about a condensed or complex nervous system. Did such a nervous system evolve separately, in each of these phyla? Or the last ancestor of the animals with bilateral symmetry had already had a strongly condensed nervous system which became dorsal in vertebrates' ancestor from its ventral position.

Willing to answer to this question, DETLEV ARENDT (1980 and 2006) focused on a complex of architect genes of the nervous system, named “median-lateral regionalization”. In vertebrates, these genes are the origin of the differentiation of the motor neurons from the sides of the spine (in its dorsal half) and of the sensorial neurons, in a more lateral position. DETLEV ARENDT studied this phenomenon in *Platynereis dumerilii* and in 2006 presented two things: - *Platynereis* has homologies in the entire medio-lateral regionalization genes of vertebrates; - these genes express themselves in the same order as in vertebrates, but in a ventral position, as a mirror image.

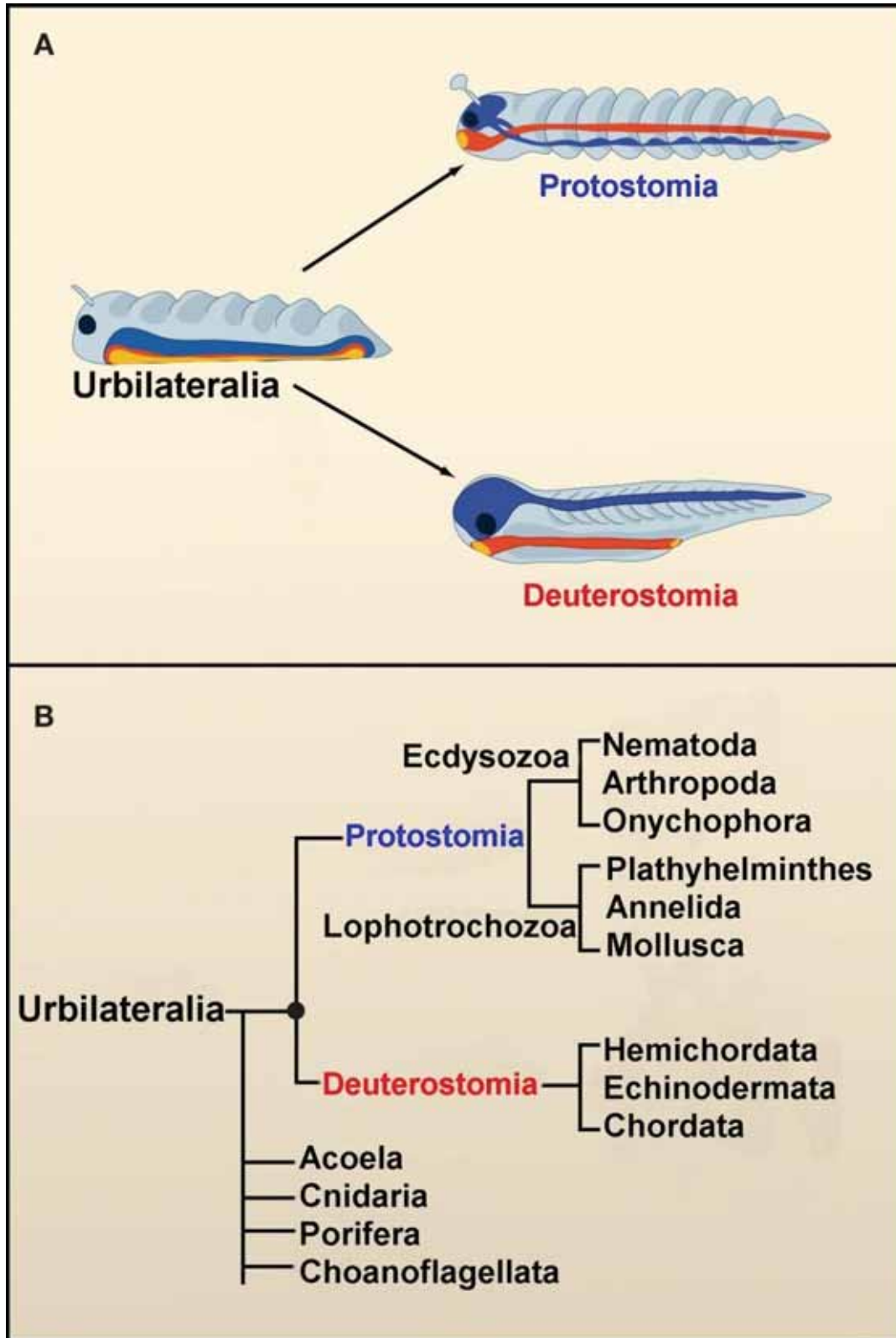


Figure 1. Sketch of the origin of animals with bilateral symmetry from a hypothetical ancestor (A) and their classification (B). / Figura 1. Schița originii animalelor cu simetrie bilaterală dintr-un strămoș ipotetic (A) și clasificarea lor (B) (după ARENDT, 2006, modificată).

Following the same argumentation as for *Hox* genes, it is difficult to imagine that the genetic system, so complex and so similar could appear independent in annelids and in vertebrates. Also, these similarities are a strong clue of the presence of a ventral condensed nervous system in the ancestor of the animals with bilateral symmetry (which became dorsal in the ancestor of vertebrates, in all likelihood by a rotation of the body axis).

The assembly of these studies make us think that the last common ancestor of the animals with bilateral symmetry **had a relatively complex anatomy**, especially a body formed of identical segments, a ventral condensed nervous system, a digestive track with mouth and anus, and to all appearances, simple photoreceptor organs. It cannot be asserted that they are similar exteriorly. As it regards the simplicity of the current acoelomates, this resulted in loss of some features and some associated genes, along the evolution, by an evolutionary simplification phenomenon.

But not everybody agrees with this perspective of the similarities between gene complexes from remote species. The palaeontologists DOUGLAS ERWIN, S. A. BOWRING and Y. G. JIN (2002) from the National Museum of Natural History of Washington D. C., as well as ERWIN DOUGLAS with the biologist ERIK DAVIDSON from the Institute of Technology of California asserted, in 2002, that the architect genes, present in the common ancestor of the animals with bilateral symmetry would not have assumed the functions which these genes have today, of “building” the organs. For them, architect genes of the ancestor of the animals with bilateral symmetry regulated only the differences between certain cells. Then, along the evolution, these genes were progressively used in developing of the more complex organs, including these ancestral cells.

In this respect, an example is that of sight, in which the architect genes gave a photoreceptor capacity to some cells, without which the animal could not have had a photoreceptor organ, be it as simple. Then, these genes were used in the development of the complex structures, including these photoreceptor cells, this thing leading to the very diverse structures of the current animal eye.

Therefore, ERWIN and DAVIDSON (2002), proposed an ancestor of animals with bilateral symmetry, which might have had a complex genome as ours, but with the difference that it was not so complicated anatomically: a small sized worm already has some different cell types (e.g. a digestive system, sensorial cells, etc.), but still it hasn't a system of sophisticated organs. Although this idea was released in 2002, today, due to the recent results got from annelids, it is fully confirmed. But the ideal for the final cutting of these debates indubitably would be the discovery of the Precambrian fossils.

Generally, animal phylogeny has long been controversial. Within the pre evolutionary era, since the 17<sup>th</sup> century, CUVIER distinguished four phyla: vertebrates, molluscs, articulates and radiates. Soon after that, it was established that the CUVIER'S radiates (coelenterates and echinoderms) were an artificial association, later the other phyla of him being regrouped. Finally, the multicellular animals were classified in 30 – 35 distinct phyla. They referred to sponges, coelenterates, echinoderms, arthropods, annelids, molluscs, flat worms and chordates – each of them with lower phyla. Only in 1859, the evolutionists began to identify the relationships between these phyla and how they can be arranged in a single phylogenetic tree.

But a question always comes back – how did the multicellular animals look like and which of the upper taxa gave birth to other taxa, superior to them? That is why, the delimitation of the taxa basing on their resemblance had to be adopted, and not according to the ramification point criteria along time.

If all these phyla occurred in the late Precambrian and in the early Cambrian (about 565 – 530 million years ago), the issue of the lack of intermediate fossil phyla remains, as well as the lack of intermediate living organisms. That is why phyla seem to be separated by unbridgeable gaps.

But how are the gaps explained?

Primitive animals did not leave fossils. Therefore, phylogeny has to be reconstructed by the study of their living descendants. Careful comparison of the invertebrate morphology and embryology led to an acceptable solid construction, after 100 years, which represents the phylogenetic tree of animals. Convergence, parallel evolution, extreme specialization, mosaic evolution, loss of the important features and other evolutionary phenomena seemed to prevent further progress, for a while. This impasse was broken when molecular characteristics were added to morphological evidence.

When it was discovered that the molecules which form the genes evolve and have a phylogeny as the morphological features, they hoped that one can build an accurate phylogeny of organisms. Molecular evidence had to allow decisions every time when morphological data were unclear. But things proved not to be so simple, because the phenomenon of mosaic evolution was ignored. Each component of the genotype may evolve independently by the rest of the genotype in a certain extent. The attempts to construct phylogenetic trees based on changes in a particular molecule often led to solutions which were clearly inconsistent with a large amount of morphological or other evidence.

Because of technical reasons, the used molecules for the first time in such analyses were the ribosomal RNA and mitochondrial DNA. Unfortunately, these molecules often have their own evolutionary path. Especially the phylogenies based on RNA 18S milled. In all recent molecular analyses conclusions are based on the study on several molecules, including nuclear genes.

But, some occasional failures do not diminish the extraordinary contribution of molecular evidence. These new evidence allow us now to build well verified phylogenies of the animal kingdom, initially made only basing on the remarkable achievements of morphology and embryology.

The most primitive living multicellular animal is *Trichoplax* (**Placozoa**) which is formed of a ventral layer of cells and a dorsal one. It reproduces by "swarming" or division. It follows the next taxon, the sponges (**Porifera**), whose ancestors seemed to be choanomonads. Molecular analysis suggests that the coelenterates, the next step in the animal evolution derived from sponges. It might be possible that coelenterates occurred independently from other group of protists. The two coelenterate phyla (**Cnidaria** and **Ctenophora**) have morphology with radial symmetry. Their embryos have two cellular layers, an ectoderm and an endoderm, i.e. they are **diploblastic**. All other multicellular animals (**Bilateria**) have a bilateral symmetry and the third cellular layer - mesoderm, i.e. they are **triploblastic**.

From now on we can talk about the biodiversity evolution in the meaning of Darwin's theory.

Before using molecular analysis, chosen classification entirely depended on the importance given to different morphological features. The presence or the absence of the coelom was (wrongly) considered as the most important feature, for a long time. Acoelomate **Platyhelminthes** were considered the base group of the bilaterals. Today, it is also accepted an alternative point of view, according to which **Platyhelminthes** are a derived group, which lost both coelom and anus, along its evolution.

All the representatives of the **Bilateria** group have a soft body; the most primitive of them crawl on the bottom of oceans or of other waters. Other primitive representatives can make tunnels through the substratum, not only for protection but also for exploring the rich source of food, available in this niche. Peristaltic contractions of a muscle layer strongly mesodermic allow putting pressure on soft substrate. This propulsion is possible by the compression of the muscles of the body walls on the cavities from the body, full of liquids. In some phyla, blood from tissues is used as propulsion liquid. Special cavities, full of liquid (so called coelom) are present in the structure of many other organisms. This hydrostatic system, which is formed of the muscles of the body walls and coelom, gives necessary rigidity to peristaltic locomotion.

**Protostomia** and **Deuterostomia** – represent the division of the **Bilateria** group into two evolutionary lines. The blastopore in the gastrula stage of an embryo of a protostomian (in developing stage) becomes the oral opening of the adult, and the anus is newly formed at the end of the gastrular bag. In a deuterostomian, permanent mouth is a newly formed opening, and the blastopore becomes anus. In addition, these two branches of animals are also different in the way in which the coelom forms.

Annelids, mollusks, arthropods and a large number of lower phyla form the protostomians group, while echinoderms and chordates (including vertebrates), together with three lower phyla form the deuterostomians group.

We recall the main differences between the two lines of bilateral organisms:

- In most of the protostomians, the egg segmentation is made in spiral, in which the plane of cell division is oblique to the vertical axis of the embryo;
- In deuterostomians the egg segmentation is radial.

But we have to be aware that some protostomians develop by radial cleavage. But the final function of each part of the zygote is **definite** from the beginning; segmentation in deuterostomians is **indefinite** – i.e. cells resulting from first division maintain their capacity to develop into a complete embryo.

Taking into consideration the morphological features, controversy arises on the phyla which have to be assigned to the protostomians and which ones are assigned to deuterostomians. Probably the subdivision of the protostomians is more difficult, with their numerous phyla. But, using the molecular analyses, things simplified much more. Mathematical methods allowed molecular information transfer in branching points of the phyletic lines. It is about cladistic analysis or genealogical, according to which only the derived features provide useful information for finding the branching points.

Usually, 24 phyla are recognized in protostomians. It was not established whether lower taxa (**Pogonophora**, **Echiurida**, **Micrognathozoa**) deserve the rank of phylum or rather should be considered classes or subphyla. Application of molecular methods showed that some complex characters (segmentation, coelom, spiral cleavage of eggs, presence of trocophore larvae) are not decisive evidence of relatedness, as always assumed, because they can be lost during evolution. For instance, the ancestors of the molluscs and pogonophorans were segmented, and those of the platyhelminthes have a coelom. But all descend from a common ancestor!

As it regards the chronology of the evolution of plants and animals, we can assert that the oldest animal fossils originate in the late Precambrian (550 million years). It was established that the massive distribution of the animals was during a very short period of time, of only 10 – 20 million of years – which is not true.

Today it is widely accepted that, in the beginning, life was only in water. The first land plants date from 450 million years, and the first angiosperms – from the Triassic (200 million years). Insects (today, the dominant group) occurred 380 million years ago. Although chordates occurred 600 million years ago, land vertebrates (amphibians) were found, for the first time, in 460 million years old strata. Soon they gave rise to reptiles, and later (over 200 million years) and from the latter resulting the birds and mammals.

Each geological era is characterized by the development or by extinction of some groups of organisms. The Cambrian, which began 543 million years ago, is the era of the first important development of the multicellular eukaryotes. The entire previous history of the Earth is known as the Precambrian (4.6 billion - 543 million years). In the first part I show that for at least a billion years from the date deducted after life began deducted for the emergence of life (with 3.8 billion years ago) **only prokaryotes existed**.

However, at a time in the Proterozoic era (2.7 – 1.7 billion years) simple eukaryotes occurred and, soon after that, - the first multicellular eukaryotes. Even if they did not left fossils, the first date of their occurrence can be deduced from the advanced evolution of their descendants from the Cambrian and with the calculation of the “molecular or evolutionary clock”. Ediacara fauna, from the end of the Precambrian (650 – 543 million years ago) is **the first animal fossil fauna**.

The period rich in fossils from the Cambrian till now is named **Phanerozoic era**. It is divided in the Palaeozoic, Mesozoic and Cenozoic eras – each of them with shorter periods. The limit between the Palaeozoic and the Mesozoic, more precisely the end of the Permian, was marked by a mass extinction of the creatures. Then, the limit between Mesozoic and the Cenozoic (at the end of the Cretaceous era) was also marked by another mass extinction, as the classic case of the dinosaurs.

Table 1. Times of occurrence of the main classes of vertebrate. / Tabel 1. Vechimea apariției diferitelor clase de vertebrate.

Vertebrate Class	Period	Occurrence Time
Gnathostome fish	Ordovician – present	450 million years
Sarcopterigian fish	Silurian – present	410 million years
Amphibians	Devonian superior – present	370 million years
Reptiles	Pennsylvanian superior – present	310 million years
Birds	Triassic superior – present	225 million years
Mammals	Triassic superior – present	225 million years

Living land plants (moss fossils) were found from the Devonian period, but certainly moss existed earlier than the fossils were left. They evolved from the charophycean algae. Symbiont fungi played an important role in the conquest of land, then inhospitable. The fossils of the first vascular plants date from the Silurian. Prevalent plants of the Palaeozoic Era (especially in Carboniferous) were lycophytes, ferns and seed ferns.

Mesozoic was dominated by gymnosperms (especially by cycadaceae and conifers), and the angiosperms, occurred in the Triassic, did not developed till the Cretaceous (125 million years). Till present, about 270,000 plant species were described, classified in 83 orders and 380 families. By the combination of the morphologic and molecular methods, the relationships of the angiosperm orders are well known. Great distribution of the flower plants took place in the middle of the Cretaceous, together with a similar huge distribution of the insects.

In order to explain the remote origin of the vertebrates, we have to make a trip just to choanoflagellate group (between protists) of which the sponges originated (**Porifera** – the most simple animals), then the diploblastic coelenterates (**Cnidaria**, **Ctenophora**), followed by triploblastic **Bilateralia**, the last ones being divided in **Protostomia** and **Deuterostomia**.

Deuterostomes are structured in four phyla: **Echinodermata**, **Hemichordata**, **Urochordata**, **Chordata**. *Amphioxus* – one of the oldest chordates survives and almost looks like our most primitive ancestors. Because it still has gill slits and a dorsal notochord, *Amphioxus* is classified with vertebrates, in Phylum **Chordata**. It feeds filtering water, but it was presumed that the most primitive vertebrates were predators. A vertebrate class strongly related is that of the extinct conodonts, which had an elaborate set of strong teeth, found in abundance in the fossil layers. The most primitive fossil vertebrates dates for 530 million years (in Yunnan – China) and belong to a fish. Jawless fish (myxiniids and lampreys) were found as fossils, old of 520 million years, and the most primitive vertebrates with teeth (placoderms) are vanished. In the above table the occurrence time of the other classes of vertebrates are marked.

Whether biodiversity refers to the entire range of variability of the creatures from a certain ecologic complex, the aquatic and terrestrial palaeo-environments were complexes which offered critical periods as well as prosperous ones for the living creatures, on their journey of becoming over until today, when we characterize them from the perspective of the genetic, ecosystemic and specific diversity. Today we try to arrange them in groups as natural as possible, groups which in turn succeed in an order imposed by many millions and billions of years, on the long becoming road of the genetic, ecologic and specific diversity.

At the same time, humanity faces climate change signals, caused largely by the unprecedented rhythm of deforestation, industrial CO<sub>2</sub> emissions in the atmosphere, fragmentation and habitat destruction, which together with the increasing exploitation of natural resources and the restriction of natural ecosystems destruction drew the threat of extinction of 1/3 of the species of gymnosperms (conifers being severely threatened), of 1/3 of the amphibian species, of 1/5 of mammal species etc. Biodiversity Conservation Convention gathers around 200 countries of the world, which proposed programs to reduce extinction rates, among others, by extension of protected areas (Natura 2000), and the United Nations Organization declared 2010 – **International Year of Biodiversity**, marked by a summit in New York.

## CONCLUSIONS

1. Biodiversity (specific, ecologic, genetic) is an attribute of life.
2. 3.8 billion years ago, abiogenic syntheses of the macromolecular polymers and complexes capable of metabolism met the main features of living, without acquiring the procuring method of energy and to reproduce.
3. For 1 billion years, prokaryotes (cyanobacteria, gram-negative bacteria, gram-positive bacteria, purple bacteria, archaeobacteria) were the single form of life which however could change genes between them.

4. The first eukaryotes occurred from a symbiosis of an archaebacterium with an eubacterium. Therefore, the new taxon (Eukarya) combines both the features of the archaebacteria and of the eubacteria.

5. Eukaryotes spectacularly diversified after their occurrence, about 2.7 billion years ago, resulting 36 protist phyla: amoebae, microsporidians, slimemucilaginous molds, dinoflagellates, ciliates, sporozoans, cryptomonads, flagellates, xanthophytes, diatoms, brown algae (some of them multicellular), oomycetes, myxosporidea (Myxozoa), red algae, green algae, radiolarians, to whom there are added other 20 phyla, with less taxa.

6. In the Cambrian, a real explosion of protists produced, this time proved by the presence of numerous microfossils.

7. Along the biodiversity evolution, multicellularity occurred repeatedly. Even among bacteria there were many ancestors of the multicellular organisms. The first step to multicellularity was the association of the unicellular eukaryotes. An increase in size of the cell aggregates followed, which led to a labour division in those “aggregates”, then associated in an authentic multicellular organism.

8. The first living multicellular animal (identified by fossils) was *Trichoplax* (taxon **Placozoa**) which is formed of a ventral cell layer and a dorsal one. It reproduced by “swarming” or division.

9. The next taxon (of sponges - **Porifera**) had choanomonads as ancestors.

10. Molecular analysis suggests that the coelenterates – the next step in animal evolution, derived from sponges. (It is not yet excluded the independent occurrence from some protist groups).

11. The two phyla of coelenterates (**Cnidaria** and **Ctenophora**) have a morphology with radial symmetry. Their embryos have two cell layers, an ectoderm and an endoderm, i.e. they are **diploblastic**.

12. All the other multicellular animals (**Bilateria**) have a bilateral symmetry and the third cellular layer – mesoderm, i.e. they **triploblastic**.

13. Further on, biodiversity evolution developed along the geological eras (with crisis periods, mentioned in the text) basing on the principles and under the influence of the factors invoked by DARWIN’S theory.

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