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Hypothesis

It Takes Two to Evolve Too.

The Hypothesis on Three Primary Communication Transitions in Evolution

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Abstract: Currently there is little doubt left on the symbiogenetic nature of eukaryotes - genomes of archaeon and bacterium participated in shaping a genome of last eukaryotic common ancestor in equal albeit asymmetric manner, while a merger event itself indicated the advent of a new domain of life. The "symbiogenetic" framework of interaction of two partners is proposed, outlining similar steps essential for three major advents: the origin of life, the origin of complex life and the origin of humans. Given the immense importance of proper energy source for the evolution of life it seems plausible that for any principal increase in complexity, a partnership with a novel energy donor is required. Moreover, a "language" elaborated in the course of communication of partners might have been a prerequisite for a subsequent increase in complexity. Transitions, which led to RNA-protein world, eukaryotes and human brain, resulted in advent of complex languages via communication onsets between two entities in close partnership. Accordingly this further facilitated formation of first cells, multicellular organisms and human society.

Keywords: symbiogenesis; eukaryogenesis; RNA world; expensive brain; human evolution; hydrothermal vent

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1. Introduction

It is intuitively clear that gradual increase of complexity was present in the course of evolution, however it is not that clear how to measure this complexity. Several major stages in the evolution of complexity and the transitions between them have been proposed in [1,2] and revisited in [3-5]. In different versions seven to eight transitions have been proposed, corresponding to the origin of protocells, chromosomes, genetic code, eukaryotes, sexual reproduction, multicellularity, eusocial animals and human societies. Here I argue that there have been three primary transitions (see fig. 1), each resulted in a novel "communication onset", while other major transitions from the list might have been predetermined based on former three (see 3.1). The first transition took place at the pre-life stage of evolutionary events and coincided with the advent of translation. I suggest that once a protein analogous to ATP synthase fortuitously evolved, it tremendously increase an energy supply for the set of compartmentalized replicators. New source of energy allowed the advent of communication of two major biopolymers on Earth and "invention" of gene expression - protein enzymes. This in turn opened the unlimited possibility of division of labor [1] between amino-acid-based enzymes and nucleic-acid-based genes, which almost inevitably led to the formation of first cells. The second transition is eukaryogenesis, which started as a partnership of α-proteobacterium and archaeon. I suggest that after α-proteobacterium becomes an energy donor, during the probable phase of conflict resolution between two partners, the new way of communication had been arisen and elaborate; the one where genes from one entity using different physico-chemical signals,

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including signaling molecules enclosed in vesicles, communicate to genes from another entity. The onset of third communication – human language and culture - coincides with the advent of human consciousness and intelligence and one can only hypothesize what are the causal connections between these major steps in human evolution. I argue that additional energy, provided by paternal care to female and offspring made human encephalization possible and resulted in a successful division of labor between hominid males and females. This advent of a primary unit of society facilitated the evolution of human mind capable of a novel form of complex communication.

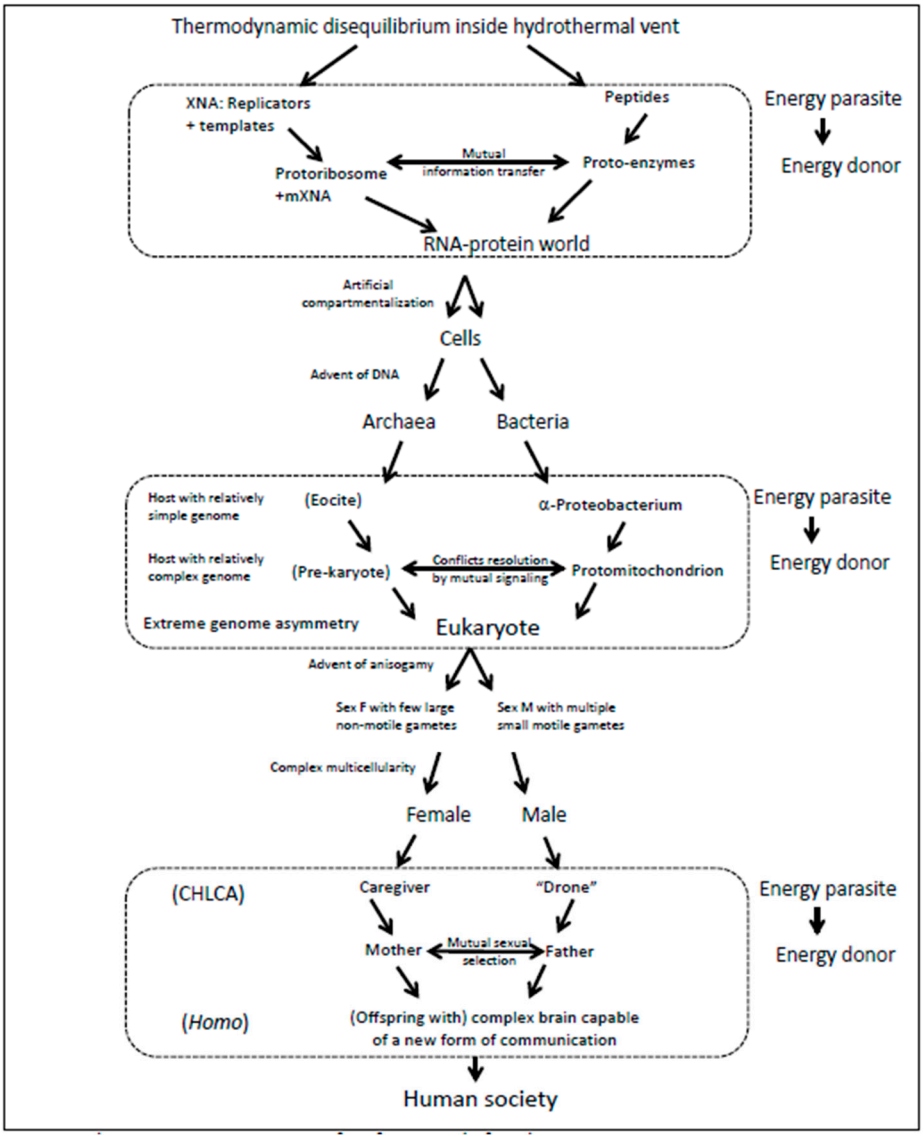

Fig. 1. Schematic representation of a framework for three primary communication transitions (boxed). During the first step of each of transitions an energy-parasitic partner turned into an energy donor. During the second step, accompanied by mutual informational transfer, elaborate "language" evolved due to communication between two partners. XNA – nucleic acids with unknown backbone, CHLCA - chimpanzee/human last common ancestor.

2. Three primary communication transitions

Communication, as determined by E. O. Wilson in his Sociobiology [6], is action on the part of one organism (or cell) that alters the probability pattern of behavior in another organism (or cell) in an adaptive fashion. Zahavi [7] mentioned that the only systems, having signals that do not invest in reliability are human language and genetic code (and I add here cellular signaling in multicellular organisms). Hence apparently division of labor between entities with the same interests predisposes

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them to the evolution of fair communication without unnecessary costs. Here I would like to uncover the similar mechanism of evolution of these communications by fine-tuning signaling systems between energy parasite turned into donor and its beneficiary. First I "set the sages" and describe the "characters" (2.1 – 2.3), and later (2.4) propose the general framework for the three communication transitions.

2.1 The first transition: RNA-protein world

Several hypotheses exist on the origin of the first pre-life forms. The major dilemma is between metabolism-first and information-first approaches. Among the former hypotheses unequivocally the strongest one is the hydrothermal vent hypothesis [8], while proponents of replication-first approach almost unanimously support the RNA World hypothesis [9-11] (with some uncertainties recently regarding the first backbone, hence I'll call it the XNA World). The weakest part in the latter hypothesis (as well as in all replication-first hypotheses) however is the absence of stable sources of energy and monomers required for the polymerization. Currently the absolute majority of energy needed for biomass growth on Earth is coming from photosynthesis – the solar energy. However it is known, that photosynthesis is a late bacteria invention and the corresponding genes thus were not present in LUCA (Last Universal Common Ancestor of all cells) [12]. Hence the first pre-life forms must have been chemoautotrophs. The protein, universally present in both Bacteria and Archaea and which gene was certainly present in LUCA is primordial homolog of ATP synthase – protein, generating ATP by utilizing transmembrane chemiosmotic gradients. It has been proposed first by M. Russel and elaborated further by Russel, Martin, Lane and others [8,13-15] that natural proton gradients and FeS minerals inside submarine hydrothermal vent systems have driven the origin of life. The above mentioned original papers and reviews contain excellent and detailed description of this highly plausible hypothesis, so I just briefly mention here the main assumptions related to the following communication transition. The process of serpentinization (olivine at ocean crust reacts with water to form sertpentinite and H_2) at alkaline hydrothermal vents generates natural proton gradients of the magnitude and orientation used by modern cells. Thermally driven convection of ocean water brings CO_2, which together with H_2 are two major food resources for the proposed first chemolithoauthotrophic cells – archaeal methanogens and bacterial acetogens [16]. With the help of semiconductor FeS barrier, which separates alkaline hydrothermal fluid (pH of 9-10) from ocean (pH of 5.5-6) electrons transfer from H_2 at alkaline pH to CO_2 at pH 6 and simple organic molecules can be formed. Hence hydrothermal vent might have provided the source of food and the source of energy, reflecting current driving force of ATP synthesis. Meanwhile thin FeS mineral layers might have formed membrane precursors responsible for separation internal vent compartments from each other and from an outside ocean.

The formation of complex organics, including nucleic acids components under plausible hydrothermal vent conditions described in details elsewhere [17]. The XNA World (where nucleic acids with yet unknown backbone performed informational and catalytic roles) takeover might have started with hypercycles described in [1,18]. In this scenario, several replicators are united in a cycle, where each preceding member facilitates replication of succeeding one. The simplest scenario can be imagined, where two replicases are complementary strands of one nucleic acid molecule.

Ribosomes are present in all living organisms and represent the fundamental hallmark of cellular life (in contrast to even largest viruses). Ribosomal RNAs were unequivocally among the first and most important molecules in mixed RNA-protein world and the origin of rRNAs should be tightly connected to the origin of genetic code/translation. There are two alphabets currently present in biopolymers – nucleic acids and amino acids (aas) - with a ribosome literally translating one into the other with the help of the genetic code. The question of how genetic code emerged has no definite answer yet, and various existing hypotheses can be found in several publications [19-22]. One particularly interesting hypothesis [23] suggests that protoribosome originated in an RNA world as a replicase that added trinucleotides (which had been cut out from tRNA-precursor) to the growing strand of RNA. Somewhat modified hypotheses [24,25] suggest that this energetically unfavorable

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process can be accelerated by coupling it to energetically favorable process, such as the formation of peptide bonds in growing protein attached to pre-tRNA in similar way, as it is happening in the modern ribosome. Hence in this model applied to XNA world (Fig. 2a), peptide bonds formation would be coupled to "triplication" process and the formed peptide would be a parasitic byproduct of XNA-dependent XNA-performed XNA polymerization. But what was the mechanism of choice of particular aa "handle" [26], attached to the 3'-end of tRNA precursor? Unlikely it was random, the idea of stereochemical affinity between aa and the anticodon part in the pre-tRNA is much more plausible. As reviewed in [20], based on aptamers screening [27], several aminoacids have strong affinity towards their corresponding anticodon nucleotides, especially 2 and 3 that corresponds to the most critical in the process of translation codon nucleotides 1 and 2. Authors make a conclusion that these interactions possibly played a role prior to the establishment of translation. Moreover authors show a clear path for transfer of just 7-nts long pre-tRNA into modern clover leaf tRNA structure. Thus charged pre-tRNA (Fig. 2b) might look like 5'-$Z_1Z_2Z_3$DCCA-aa_1, where $Z_1Z_2Z_3$ are first, second and third nucleotides of anticodon corresponding to aa_1, while D - a coding base-determinator together with CCA form a consensus 3'-end of a modern tRNA [20]. Hence in a pre-translation world, amino acids might have been charged to a pre-tRNA in a semiselective manner due to an amino acid's stereochemical affinity to a corresponding anticodon, possibly using DCCA sequence as a nucleotide linker. If the interaction of particular aa with anticodon depends mostly on atoms of nucleobases, I argue that this way stereochemical affinitiy of aa to anticodon might not depend much on a backbone, rather can be quite similar among any XNA. The importance of such amino acid handle, besides facilitating replication, is discussed in 3.2 – briefly achiral nucleic acids adopt right handed form due to chiral induction [28] by terminal L-amino acid residue.

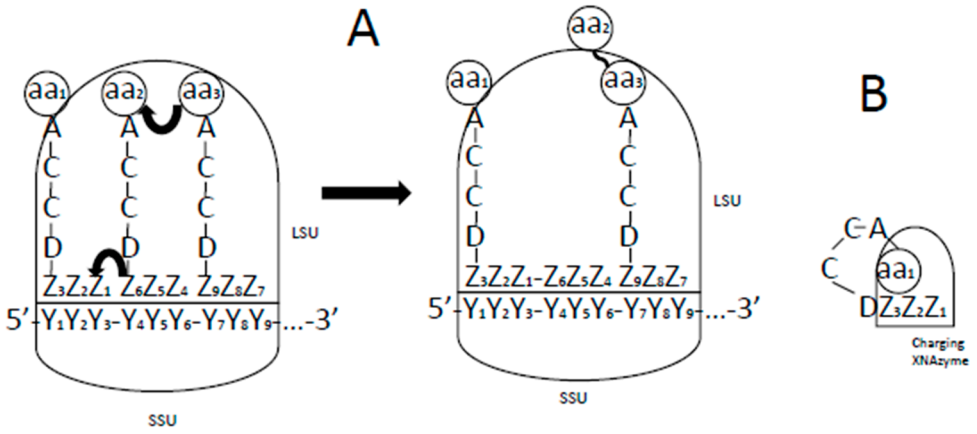

Fig. 2. A. Scheme showing the formation of peptide bonds during the process of replication of template strand, represented by codon triplets (Y_{i-1} Y_i Y_{i+1}), by the protoribosome (triplicase), represented by two subunits (SSU and LSU). Peptide bond formation between aa_2 and aa_3 (while aa_1 is permanently linked to the 3'-end of a growing strand) is coupled to the ligation of the first anticodon triplet ($Z_{i-1}Z_i$ Z_{i+1}) to the second one. B. Charging of 7-nt long pre-tRNA with amino acid mainly based on affinity of aa to the second and the third nucleotides in the anticodon by hypothetical XNAzyme.

Peptides synthesized this way were waste products, parasitizing on a triplicase, and because of the absence of a strict code were highly mutable. Let's imagine that once a protein was accidentally synthesized that was able to significantly help a set of replicators in the same compartment with a gain of energy, say by direct transforming of natural proton gradient into high-energy bond formation, reflecting the function of nowadays ubiquitous ATP synthase. This innovation should incredibly increase the fitness of this compartmentalized set of replicators. However to keep this evolutionary advantage, replicators would likely had to at least partially fix a genetic code or even adjust their own structure. If this is correct, then translation has begun with the informational flow Peptides ⟶ Replicators directed the opposite way from that stated in the Central Dogma.

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Before the invention of the first phenotype a very limited pool of replicators – templates had been compartmentalized, but once an additional stable source of energy and a genetic code emerged, additional replicators – genes might have evolved, moreover selection pressure mostly switched to the phenotype level. This allowed the selection of efficient protein-based XNA-dependent XNA polymerase, while protoribosome was freed from a triplicase duty and turned into a protein-synthesis machine. Furthemore, virtually any XNA molecule, uptaken from the neighboring compartment - primordial Horizontal Gene Transfer – HGT - might have turned into a gene and even a slight display of any useful function of the correspondingly translated protein followed by mutations and selection, had resulted in the emergence of a new enzyme. Following this process of initial gene pool formation one can easily imagine the advent of both selection-favored less error-prone nucleic acids with novel backbones and artificial compartmentalization.

2.2. The second transition: Symbiogenesis

As mentioned above, the first archaea were most likely methanogens, while bacteria – acetogens [16] and this separation probably have started before the advent of cells. But the presence of much more complex eukaryotic cells along with these two types of simple prokaryotic cells is yet to be explained in details. There have been numerous theories of the origin of eukaryotic cells which have been recently reviewed excellently [29-31]. Genome complexity and chimerism, endomembrane system, cytoskeleton, presence of various organelles with own genomes – that is not a complete list of eukaryotic hallmarks, that have until recently never been either explained to emerge *de novo* in eukaryotes or found existed in prokaryotes. Despite of consensus among biologists that eukaryotic cell is a product of symbiosis of some archaeal relative and α-proteobacterium, currently no theory of eukaryogenesis, explaining all eukaryotic hallmarks, is widely accepted. It is accepted, however, due to unequivocal phylogenetic data, that α-proteobacterial partner gave rise to mitochondria by genome streamlining and massive gene transfer to nuclear (archaeal) genome.

One critical observation, related to the genome complexity of eukaryotes, which has recently been made by Lane and Martin [32-34], points to the fact that eukaryotic protists and bacteria generate nearly equal energy per megabase of DNA, in contrast to much greater requirements of energy per gene in eukaryotes. Hence this strongly supports symbiogenetic scenario of eukaryogenesis by indicating that in the absence of energy-generating mitochondria (which have been acquired relatively late), it would be impossible for eukaryote to support its enormously large genome. Another recent critical discovery is identification by metagenomic analysis of a new candidate archaeal phylum – Lokiarchaeota [35], which genome possesses many eukaryotic hallmark genes, including crenactins (archaeal analogs of actin), ESCRT pathway, ubiquitin system and some others. These data suggest that perhaps host cell was a *bona fide* arhaeon, albeit relatively complex, confirming symbiogenetic eukaryogenesis scenario. Altogether symbiogenetic hypotheses, for instance "ring of life" [36] hypothesis, suggest a merger scenario of eukaryogenesis, where genomes of archaeon and bacterium participated in shaping a genome of last eukaryotic common ancestor in equal albeit asymmetric manner. This hypothesis correlates well with the complex genome advent following energy requirements fulfilled by mitochondria "powerhouse". Hence eukaryotes form a secondary domain of life derived from two primary prokaryotic ones. Philogenetic studies of chimeric eukaryotic genome [37] also fit this hypothesis.

Regardless of particular hypothesis, phylogenetic trees show that *Lokiarchaea* [35] are the closest archaeal relatives of eukaryotes, yet data available on Lokiarchaeota are based on metagenomic sequencing analysis only and no species have been ecologically or physically described. But let's take a look on another very closely related archaeal phylum Thaumarchaeota [38], which also possess ESCRT pathway. It seems to be intrinsically predisposed to symbiotic partnerships: among very few described species two are involved in symbiotic relationships – *Cenarchaeum symbiosum* [39] is a symbiont of marine sponge, while a giant cells of Candidatus *Giganthauma karukerense* [40] are covered with multiple smaller γ-*Proteobacteria* symbionts.

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While some of the hypotheses on the mitochondrial origins posit mutually beneficial association with host (e.g. syntrophy [41] and [42]), I suggest that the initial nature of α-proteobacterial association with archaeon, which led to symbiogenesis, was parasitic-host. α-proteobacterium might have been an exoparasite and have been engulfed at a later stage, or endoparasite, if archaeon possessed phagocytosis ability. Parasitic nature of protomitochondrion has been proposed multiple times (for instance see [43,44]). It is not unlikely that α-proteobacterium, resulted in a mitochondrion had been primarily an archaeal parasite. The bacterial order Ricketsiales where mitochondrion is currently placed based on phylogenetic studies [45], contains several obligate intracellular parasites, including members of genera *Anaplasma*, *Rickettsia* and *Wolbachia*. Parasitic way of life should favor the emergence of basic signaling pathways. Moreover massive gene loss is typical for parasitic bacteria. Furthermore, recently it has been predicted [46] by phylogenomic reconstruction that mitochondrial ancestor possessed parasite type of ATP translocase, which imported ATP from the host. Thus the parasitic origin of mitochondrion seems plausible. But what could have forced protomitochondrion to turn into mutualistic organelle and reverse the flow of ATP? It is known that vertical transmission tightly interconnects the interests of hosts and endosymbionts. Accordingly, transfer from horizontal to vertical transmission of protomitochondrion might have enhanced its mutualism and cooperation. The last step required an extreme adaptation of partners to each other and moderation of conflicts (see 2.4) probably through the onset of a new type of communication – signaling molecules enclosed in membrane vesicles, transported by cytoskeletal systems. The latter systems were evolved or further developed during symbiogenesis due to an increasing amount of energy provided by mitochondria. Gradual decrease of mitochondrial genome and increasing number of mitochondria allowed support of more and more complex nuclear genome because of extreme genomic asymmetry [34]. Lane [34] approximated the host energetic benefits of losing just 5% of protomitochondria genes. It turns out that it would save about 580,000 ATP molecules per second. These benefits could be transferred into developing costly traits like dynamic cytoskeleton (580,000 ATP roughly equals 4.5 micrometers of actin filaments growth) – supply networks that might substitute the endosymbionts' loss of essential substrates.

Some genes however are required to be expressed locally in mitochondria [47] to maintain proper control over electron flow and membrane potential. Later similar mechanisms of control over membrane potential might have been used and further developed for the maintaining of electron gradients across membranes in neurons.

2.3. The third transition: Human brain and language

It is known, that around 6 million years (Ma) ago [48], possibly after complex speciation [49], human lineage diverged from the chimpanzee/human last common ancestor (CHLCA), but the sequence of events leading to humanization is not yet known. Major traits separating humans from chimpanzee include increased brain size, bipedalism, monogamy and decreased sexual dimorphism, elongated ontogeny, loss of body fur, precision and power grip [50].

Both bipedalism and monogamy (for the recent hypotheses on the reasons of early human monogamy see [51-53]) with paternal care are likely starting points for other key changes in humans. Paternal care was likely a requirement for lengthened gestation and especially infancy - critical factors, which made human encephalization physically possible. For instance, based on the expensive brain hypothesis, advocated by K. Isler and C. Schaik [54-56], costs of larger brain in early hominids must be met by increased energy provision to female and offspring, possibly by paternal care. Likewise Kaplan [57,58] studying evolutionary roots of human social organization pointed out the essential role of males in energetics of human reproduction, in contrast to most higher primates. Their analyses of food provisioning among forager societies show that among the Ache (hunter-gatherers in Paraguay) the total expected net food production (calories produced minus calories consumed) from age 18 to death is +21,638,000 calories for men and -924,000 for women. Similar data were obtained for the Hiwi (South American hunter-gatherers): +11,151,000 calories for men and -3,096,000 for women. Hence this clearly shows that in traditional foraging societies (probably reflecting those

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of early *Homo* species) male provisioning is vital for surviving of female and offspring (whose net production is also obviously negative). The essential role of paternal care is further strengthened given decreased mobility and higher energy expenditures [59] of females carrying large infants [60] with lost ability to ride dorsally [61,62].

Power and precision grip possibly developed after the upper limbs were freed from quadrupedal movement. Precision grip was favored by natural selection, because of (heritable) tool usage which also might be responsible for the initial brain growth 2 - 1.5 Ma ago up to 600 cc (cubic centimeters) in *Homo habilis* – 800 cc in *Homo erectus* [48].

For the next million years stone tools were largely unchanged, while the growth of brain accelerated. Several hypotheses explain human encephalization by social factors (including "Machiavellian intelligence" [63] and "social brain" [64] hypotheses). However some authors, to solve this paradox, followed Darwin [65] who emphasized a role of sexual selection in human encephalization, hypothesize, that a large human brain was a direct product of sexual, rather than natural selection. One of the proponents of this idea, G. Miller [66] suggests that male and female humans mutually selected each other based on ornamented language, sense of humor and other communication means as fitness indicators. Highly developed human brain, responsible for communication skills and consuming around 20% of humans metabolic energy is in perfect agreement with a handicap principle [67], declaring that a handicap should be essentially costly to display. Here is a description of human brain by Miller [66]**:** "Sexual selection made our brains wasteful, if not wasted: it transformed a small, efficient ape-style brain into a huge, energy hungry handicap spewing out luxury behaviors like conversation, music, and art."

Although Miller describes the process he envisaged for humanization [66] as a pure sexual selection, I would rather attest it as sexual selection multiplied by natural selection, since mutually selected fittest parents should also produce fittest offspring with much higher fecundity and survival chances than that of low fitness parents. Since the emergence of strict monogamy among hominids is unlikely, pair-bonding among early *Homo* species would likely be a serial monogamy. Serial monogamy with significantly different chances of survival among offspring of parents with different fitness levels could at least partially explain seemingly polygynous behavior proposed for *Homo* species [68]. The higher survival chances of fittest offspring are due to obvious evolutionary advantages, provided by largest brain – better usage of stone tools and later language - memes *sensu* Dawkins [69].

2.4 General framework for three communication transitions

Lane in his book [70] writes about the importance of mitochondrial energy for a complex life: "…I do not believe that bacteria will ever ascend the smooth ramp to sentience, or anywhere much beyond slime, here or anywhere else in the universe." I argue that the additional energy resource is a prerequisite for the drastic changes that can be observed during each of three communication transitions. for three primary communication transitions. In a course of transitions (see Figure 1 for schematic framework), successful division of labor between partners made enormous increase of complexity of biopolymers, a cell and a brain possible; each transition resulted in an onset of a new form of elaborate language. Below I'll give details about "languages" that experienced elaboration due to mutual information transfer during three communication transitions.

The "language" of biopolymers as we can see it nowadays is based on weak non-covalent interactions (hydrogen bonds, electrostatic interactions, van der Waals attractions and hydrophobic interactions) responsible for the formation of precise secondary and tertiary structures of nucleic acids and proteins. The predictable structures formed by the definite combinations of letters in each of the two alphabets form a foundation for communication between nucleic acids and proteins. I argue that before this communication has been established the "language" of biopolymers had been plain and poor, resulting only in replication of a small set of possibly achiral nucleic acids inside natural compartments. The advent of communication and elaborate language might have been tightly interconnected with the advent of homochirality (see 3.2). Contingent choice of L-amino acids

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resulted in shift in helix equilibrium to right-handed that allowed predicted conformations to be fixed in both biopolymers. Upon obtaining new energy resource, communication developed starting with genetic code, followed by translation and protein-based XNA replication, which closed the circle. Subsequently, selection favored transition to D-ribose backbone of nucleic acids, complementing L-amino acids.

The language of cells – physico-chemical signaling – could not be developed enough among prokaryotes thus we can only see complex multicellular eukaryotes. Multicellularity *per se* (or simple multicellularity) exemplified by filaments, sheets or clusters of cells is not that uncommon [3,71] – many bacterial species can form these structures. However, despite some selective advantages provided by multicellular stage, bacteria could never evolve into complex multicellular structures. Complex multicellularity *bona fide*, characterized by the absence of the direct contact of some cells with the environment, requires elaborate signaling system and developed membrane and cytoskeletal systems for the transport of signaling molecules. In contrast to bacteria, in eukaryotes complex multicellularity has evolved independently several times: in animals, land plants, few groups of algae and fungi. What could have made eukaryotes predisposed for the formation of complex multicellular structures? I argue here that the whole communication system, which includes packaging signaling molecules into endosomes and transporting them by means of dynamic cytoskeleton, had been fine-tuned during the symbiogenesis. It has been noted [72] that conflict mediation might have been a key point in all major transitions. What could have mediated conflicts arising during the major steps in evolution of multicellular organisms? The key to this transition might have been metabolic signaling as proposed in [73]. The author suggests that the first signaling pathways might have emerged in bacteria and transferred to eukaryotic genome with the massive gene transfer from a proteobacterial symbiont to the host. In particular, the case of soluble adenylyl cyclase (sAC) pathway linked to near universal metabolic products (CO_2/HCO_3^-) has been considered. Cyclic AMP (cAMP) serves as a messenger in a variety of organisms. Six classes of adenylyl cyclases, which produce cAMP from ATP, are found in bacteria but only class III cyclases are found both in bacteria, including α-proteobacteria, and in eukaryotes. There is no data on the presence of class III ACs in archaea. The author [73] argues that if CO_2/HCO_3^-/sAC/cAMP signaling pathway modulates respiration in eukaryotes, it had played a major role in establishing a communication between a host and a protomitochondrion during eukaryogenesis. Hence, one can imagine, that the relative ease of the emergence of complex multicellularity can be explained by the fact that all the necessary mechanisms of conflict mediation had already evolved during establishment of communication in archaeal - bacterial symbiotic partnership.

STAT3 and VEGF-B signaling pathways also have been proposed [73] to be originated in bacteria and later transferred to the nuclear genome. As important regulators of mitochondrial metabolism, these pathways might have mediated conflicts in the prokaryote-to-eukaryote transition.

Epigenetic inheritance, considered [5] as one of the four major types of information, and apoptosis are hallmarks of eukaryotic complex multicellularity. The information on the origin and phylogeny of DNA methylation machinery in archaea is very limited, but it seems that bacterial machinery is overwhelmingly more developed, while archaeal machinery can be a result of HGT. Thus DNA methylation machinery could have been invented by bacteria and later transferred to nuclear genome of eukaryotes. Somewhat similar situation is with the programmed cell death pathways – almost no data on archaea, but clear indications of gene transfer from bacteria to eukaryotes [74]

Hence the cellular "language" has been elaborated only after bacteria had provided key signaling pathways and archaeon had provided (evolved *de novo*) membranes manipulation and cytoskeletal systems. During symbiogenesis coordinated action of these conduits resulted in successful resolution of conflicts and onset of communicaton between entities.

Following the advent of brain and sensory organs, new language emerged based on audio-visual-olfactory stimuli that delivered signals sent by one organism directly to another organism's brain which in turn starts an intercellular communication finalized by communication with genes inside target cells. Similarly to previous transitions, language developed by simple brains of animals

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was evolved to solve simple tasks of survival or courtship display. It was only a human brain entrapped into a positive feedback loop of culture/language benefits for sexual selection powered by natural selection that could grew into "the most complex object in the visual universe". Human brain has been configured for complex language first in the course of intimate partnership of *Homo* male and female (conflict resolution cannot be excluded either). Subsequently complex language has been utilized for communication of entities outside the primary unit of society (family) that in turn facilitated the transfer to modern human society.

3. Discussion

3.1 Other major transitions

I suggest that three primary transitions are probably necessary and sufficient for other transitions listed in [2,4] to happen. Let's consider briefly some other transitions, which have not been discussed yet.

Transition of independent replicators to chromosomes likely happened after advent of DNA, which is more suitable for storage of genetic information due to lesser mutability and greater chemical stability of deoxyribose. Accordingly evolutionary pressure for transfer of genetic material into the form of DNA, once latter being invented can be easily imagined. No new "language" has to be evolved since DNA is compatible with RNA. Selection pressure is probably responsible for the origin of the integrated genome. The chance of emergence of cheaters and "outlaws" (*sensu* Dawkins [69]) among separated selfish replicators is likely much higher than among joint cooperating replicators.

Eukaryogenesis has been certainly a much more complex process, than just obtaining an energy-generating endosymbiont by an archaeon. Following changes however might be explained as a result of one primary transition.

Despite the fact that the presence of the nucleus gives to eukaryotes their name, currently it is considered as a secondary trait. One of the most plausible hypotheses on the origin of nucleus suggests [75] that this invention had been a direct consequence of an endosymbiotic event. During the massive gene transfer from a protomitochondrion, prokaryotic group II introns invasion into the archaeal (nuclear) genome happened. Later group II introns successfully transformed into the spliceosome-dependent introns. However, a new problem subsequently had arisen: the spliceosome machinery had been much slower than translation machinery thus causing ribosome to translate pre-mRNA into defective proteins. One of the possible solutions of this problem would be a spatiotemporal separation of nascent transcripts from ribosomes and that is exactly what might have happened by the formation of nuclear membrane. This invention allowed the slow splicing of RNAs to happen inside the nucleus while ribosomes were allowed to translate matured RNAs only after the export of RNAs through nuclear pores.

Transition from asexual to sexual replication (with anisogamy) seems also to be a direct consequence of obtaining mitochondria during symbiogenesis. The alternation of haploid and diploid states – the prerequisite for the emergence of sexual replication – is favored in eukaryotic protists due to providing higher adoptability over constant environmental changes. The anisogamy is a result of conflicts inside zygote between mitochondria from cytoplasms of two gamets, which favored the selection for one large ovum exclusively containing mitochondria for future zygote as a gamete for female sex and multiple competing small mitochondria-less sperms as gametes for male sex. Moreover meiosis itself – the evolutionary benefit outweighing the two-fold cost of sex – might have originated as a response to symbiogenesis [76].

3.2 On the nature of nucleic acid partner in the first transition

Currently apart from traditionally favorite ribonucleic acid (RNA World hypothesis [9-11]), threose nucleic acids, glycol nucleic acids, protein nucleic acids [77] (PNA) and nucleic acids with some other backbones [78] are considered among potential candidates for the first replicator

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molecules. Subsequently, during evolution of life, RNA has been evolved as a molecule more suitable for both information storage and enzymatic functions. Proponents of the primarily alternative backbone of nucleic acids, including N. Hud [78], argue that there is no simple way of ribonucleotide monomers synthesis under plausible prebiotic conditions, moreover RNA tend to be notoriously unstable especially under hydrothermal vents conditions: high temperatures and alkaline pH. Taking into account a very close nature, yet with somewhat different roles of partners, participated in the other transitions, I suggest here, that PNA or another backbone with similar properties might have been the first information polymer. Below I will provide several reasons for this suggestion. Backbone of PNA consists of N-(2-aminoethyl)glycine units, to which nucleobases are attached via carbonyl methylene linkers. First, amide bonds used both for nucleotide attachment to backbone and for units' polymerization have been shown to be easily formed under prebiotic conditions and at 100°C [79]. Second, the formation of components of PNA has been shown even at 100°C [80]. Third, PNA can mimic double helix structure of the traditional nucleic acids, thus PNA is compatible with RNA and transfer from PNA World to RNA World could have gone smoothly [81]. Forth, PNA is a non-chiral polymer, hence, unlike in the case of RNA and DNA, there is no prerequisite for homochirality to form biologically active secondary and tertiary structures [81]. Moreover, a notorious problem of cross-enantiomeric inhibition of polymerization is not relevant in the case of PNA. Furthemore, equilibrium between right-and left-handed helixes existing in PNA can be shifted towards right-handed by the attachment of L-amino acid to the C-terminal base of one of the strands [81]. That might shed light on the possible role of amino acid "handles". Originally, non-chirality of the first nucleic acids prevented them from displaying high levels of biochemical activity. However, attachment of just one L-amino acid (in contrast to PNA, amino acids are always chiral, but most likely were synthesized abiotically as a racemate, hence a choice of L-enantiomer would be a contingency) might have shifted helix handedness and increased activity. It correlates well with the observed preferential stereochemical interaction between L-amino acids and D-ribose RNA [82,83]. And finally, the production of the PNA backbone monomer unit has been discovered in cyanobacteria [84] that can be a vestige of PNA World.

Thus, the first nucleic acid XNA (which does not necessary need to be exactly PNA) has to have plausible ways of precursors' synthesis under hydrothermal vents conditions; its backbone has to be non-chiral, but after ligation to (contingently chosen L-enantiomer of) amino acid, the equilibrium in XNA helix is switched (that is why "handles" are so important) toward one biologically active form, likely right-handed, providing easy shift from achiral XNA to chiral D-form RNA World.

4. Concluding remarks

Revolution in a whole genome sequencing and corresponding phylogenetic studies of newly discovered prokaryotic species during the last decade resulted in an enormous progress in disentangling events leading to eukaryogenesis. On the other hand, elaboration of hydrothermal vent hypothesis turned it into one of the most plausible theories on the origin of life. Further, new accounts on the descent of man, language and culture appeared. Based on a reviewed data, I argued that origins of life, eukaryotes and human brain can be considered in a "symbiogenetic" framework of interaction of two partners and onset of communication between partners resulted in elaborate language, facilitating further evolutionary transitions. Emergence of a novel energy donor within partnerships facilitated division of labor and further increase in complexity.

I am risking here of being accused in eukaryocentrism and anthropocentrism (see for instance [85] and later discussion on this [86,87]). I clearly realize that *Homo sapiens*, as well as eukaryotes, are entities not a tendency [88] and rewinding the tape of life might never lead to anything similar to either of those entities. However, as recently has been suggested [89,90], archaeal - bacterial "partnerships" could have been not that unique, since several might have been formed in the course of prokaryote evolution (including the one resulted in Haloarchaea [89]). Moreover it is very well might be that bacteria [91] especially in the form of communities [92] are not less complex than

complex multicellular eukaryotes. This hypothesis is an attempt of generalizing evolutionary leaps leading to the only known self-conscious species on Earth.

The above hypothesis can be tested in several ways: 1) to study the plausibility of XNA (e.g. PNA, see 3.2) synthesis under simulated hydrothermal vent conditions; 2) to study the ability (possibly using SELEX) of XNA-based "protoribosome" to perform at least to some extent the functions of nowadays ribosome; 3) to establish the partnership of *Thaumarchaeal* species with parasitic α-proteobacterial species and to track its evolution; 4) to carefully check the Haloarchaea species for the presence of symbiotic vestiges; 5) following Darwin [93], who emphasized the resemblance of natural selection and selection by man, experiments on artificial selection of smarter birds (say from genus *Corvus*) based on their communication ability can be performed (it has been shown for the first time recently [94], that artificial selection for large brain in animals resulted in their high cognitive abilities).

Hypothesis presented here can be falsified by a number of findings: 1) reappraisal of hydrothermal vent theory; 2) strong evidences that homochiral RNA was the first prebiotic nucleic acid; 3) discovery of archaezoa, that would disprove any symbiogenic scenario of eukaryogenesis; 4) strong evidences that encephalization in humans was uncoupled from (not necessarily verbal) communication skills.

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