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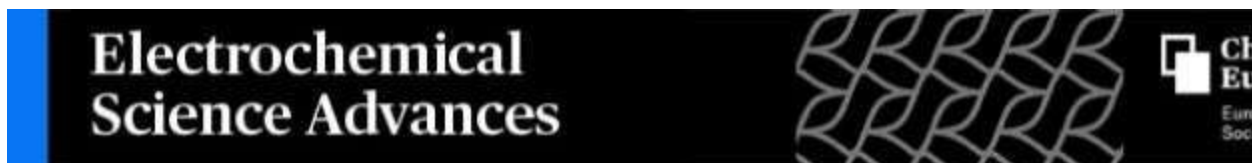
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**Aqueous electrochemistry; the toolbox for life's emergence
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Minireview

Aqueous electrochemistry; the toolbox for life's emergence from redox disequilibria

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Review Only

Abstract

The 2nd law of thermodynamics leaves no doubt that life on planet Earth and its inherent substantial decrease in entropy is fundamentally based on mechanisms converting environmental free energy into the spatial and temporal order of metabolic processes. This argument holds for present life as much as it does for its very beginnings some 4 billion years ago. In this contribution, we try to strip down free energy conversion in extant life (known as “bioenergetics” to the biologists) to its basic principles with the aim to potentially retrodict the nature of the pre-biotic precursor which drove life into existence. We demonstrate that these basic principles are deeply rooted in aqueous electrochemistry and strongly rely on inorganic redox compounds. The question of life’s emergence, generally considered to fall into the realm of organic chemistry, should therefore rather be recognized as an electrochemical problem and its ultimate elucidation will need to strongly implicate the community of electrochemical scientists.

Running title

Life’s origin relied on aqueous electrochemistry

1. A roadmap for research on the emergence of life

The bulk of research attempting to rationalize the emergence of life on our planet (and potentially elsewhere in the universe) investigates prebiotic chemical pathways producing specific organic molecules commonly associated with living matter, that is, amino acids, nucleobases, sugars and lipids, and assumes that, once plausible prebiotic mechanisms allowing the accumulation of these compounds have been found, the question of life's origin is basically solved [1,2]. As mentioned repeatedly in the past [3-5], the basic premises of this approach face substantial objections from thermodynamics and in particular from its 2nd law which stipulates that a closed system always evolves towards increasing disorder (higher entropy) and that entropy-decreasing fluctuations are the more unlikely the higher the number of degrees of freedom in the system. The 2nd law might therefore at best tolerate the spontaneous origin of living cells solely from the presence of organic molecules as an extremely unlikely "freak" accident along the lines of the "infinite monkey theorem" [6] leading Jacques Monod [7] to posit that "Man at last knows that he is alone in the unfeeling immensity of the universe, out of which he emerged only by chance" (unsurprisingly leaving the question of the whereabouts of the other half of humanity unattended).

Guided by these thermodynamic strictures we have grounded the elaboration of emergence-of-life scenarios in the assumption that life must be thermodynamically trivial, that is, predictable (at least in a stochastic manner) and ineluctable when all necessary conditions are met [8-11]. As pointed out by Erwin Schrödinger already in 1944, the apparent conflict of the order-generating phenomenon "life" with the 2nd law of thermodynamics disappears when life is understood as a subsystem of a larger entity which itself is in thermodynamic disequilibrium and thus can feed entropy decrease in the subsystem while moving the entire system towards equilibrium [12]. Free energy conversion therefore is the most crucial prerequisite to life's existence and it must have been even more so to life's emergence. The insights gained by far-from-equilibrium thermodynamics over the last half century [11,13-20] moreover show that spontaneous generation of (spatial and temporal) order in parts of the subsystem not only is admitted by the 2nd law but is straightforwardly predicted to arise in the form of so-called "dissipative structures" [21]. Rheology, atmospheric sciences and the field of chemical reactions, to name only a few, are disciplines which abound with dissipative-structure-related phenomena.

A second pillar in constructing our scenarios consists in the rigorous adherence to a top-down-approach. This means that we assume the present day functioning of living entities to closely reflect that at their beginnings and that there were no fundamental ruptures throughout evolution in the way life works [22]. This in particular concerns the mechanisms involved in free energy conversion, arguably (see above) life's most basic and essential process. We admit that, in contrast to the first one, this second assumption is arbitrary. To avoid the problem of being intrinsically non-falsifiable

[23], we try to elaborate scenarios in which life would have emerged in a form recognizable as such through the eyes of a modern biologist. This approach likely isn't shared by the entire origin-of-life community. A substantial number of articles have indeed put forward origin-of-life scenarios (in perfectly plausible geochemical settings) which produce truly alien-looking early forms of life [24-31] and which have to invoke complete remodeling of these supposedly initial manifestations of life on Earth to arrive at the observable biology of present organisms. There obviously is no way to disprove that one of these scenarios reflects the actual sequence of events. However, this is precisely what disavows them on the background of a Popperian definition of the scientific method.

The ultimate prerequisite of our approach obviously consists in a solid understanding of the unifying principles underlying free energy conversion in extant life. The elaboration of these principles (*e.g. via* the comparative study of the diversity of bioenergetic mechanisms) will then allow to search for prebiotic settings potentially providing rudimentary versions of bioenergetics, however following the same basic principles. These considerations apply both to the general types and sources of free energy and to the specific processes converting environmental free energy into the order of a living entity.

We have therefore in the past worked towards stripping down biological free energy conversion to such fundamental principles and mechanisms. Somewhat disconcertingly to the biologist, bioenergetics emerges from these assessments as a quintessentially electrochemical phenomenon with only limited (modulating/optimizing) input from the proteinaceous make-up of enzymes, but relying all-out on the extraordinary feats of electrochemical processes in aqueous (protic) media. We will in this contribution try to depict bioenergetics as what we think it fundamentally is: a network of (mainly transition-metal mediated) redox processes occurring in (and building up electric fields over) capacitor-like entities and intimately coupling redox- to protonation-reactions with the final result of generating chemical disequilibria which allow the entropy-lowering organization of living entities. Viewing bioenergetics this way suggests the possibility of a contiguous evolutionary thread linking extant life to aqueous electrochemistry in inorganic settings.

2. From extant life back to the beginnings

2.1. The entropy decrease ($-T\Delta S$) of the subsystem "Life" is driven by strong electrochemical disequilibria ($\Delta G = -nF\Delta E$) in the environment

As summarized previously [9], the universal source of free energy (ΔG) sustaining life's processes (corresponding to a lowering of entropy, $-T\Delta S$) is a redox tension between reductants and oxidants (Fig. 1). In the vast majority of cases, these redox compounds are environmental [9,32,34]. While the terminal oxidant (for the case of anoxygenic photosynthesis) or both oxidant and reductant (in oxygenic photosynthesis) may also result from photochemical reactions, substantial

phylogenetic and paleochemical evidence suggests that the photochemical generation of out-of-equilibrium redox compounds did not occur while life emerged but evolved only at a later stage [35]. This in no way should be taken to minimize the importance of photosynthesis for the evolutionary trajectory of life on Earth. It was photosynthesis and in particular its oxygenic version which freed life of the limitations of (necessarily localized) environmental redox disequilibria and allowed the harnessing of truly “extraterrestrial” free energy from the sun [36]. Colonizing the whole planet was only made possible after life had developed oxygenic photosynthesis and the emergence of complex life certainly was contingent on the enormous increase in environmental redox disequilibria engendered by the discharging of “green life’s” waste product, molecular oxygen [37].

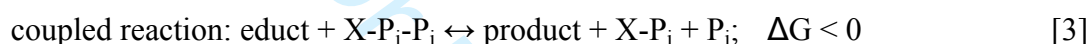
Another modification of the scheme depicted in Fig. 1 is the case of fermentation wherein reduced metabolic intermediates (such as ethanol, acetate, formate, etc) are excreted, ensuring the sustained presence of electron sinks within the respective cells [38]. Both fermentation and anoxygenic photosynthesis therefore appear as rescue mechanisms maintaining a redox tension as source of free energy when environmental oxidants become scarce or absent. Oxygenic photosynthesis fully frees life from the requirement of environmental redox tensions by using photon energy to turn ubiquitous water into a reductant and plentiful CO₂ into an oxidant allowing electron transfer reactions which would be strongly endergonic without the input of light energy [35].

2.2. What really is $-T\Delta S$ in life?

Again, a comparative analysis of the full diversity of extant life’s free energy converting systems yields a surprisingly uniform common scheme: The environmental redox disequilibria are converted into two distinct but well-defined intracellular disequilibria (Fig. 2):

(a) The out-of-equilibrium presence of strongly reducing compounds driving a host of biosynthetic processes and in autotrophs furthermore the fixation of CO₂ into biomass. In extant life, these strongly reducing compounds are either ferredoxins or NADH. Ferredoxins are essentially inorganic iron-sulphur ([4Fe-4S]-, [3Fe-4S]- or [2Fe-2S]-) clusters wrapped in a thin shell of polypeptide. NADH is the reduced form of NAD, an organic molecule the redox-active part of which is a nitrogen-containing heterocycle. The iron-sulphur clusters in ferredoxins feature midpoint potentials at pH7 in the range of -400 mV (NHE) while that of the NADH/NAD⁺ couple is at -340 mV. Both ferredoxins and NAD are present in cells at relatively high concentrations and function as “redox pools”. When high reduction ratios of these pools are attained, their operating potentials (according to the Nernst-equation) can attain values well below -500 mV for the ferredoxins and approaching -500 mV for NADH/NAD⁺. The reducing power of these intracellular reductants thus frequently greatly exceeds that of the environmental reductants [9]. Below (section 2.3.1.) we will discuss mechanisms boosting reducing power in extant life.

(2) Out-of-equilibrium concentrations of the condensed versions of the phosphate anion. The most prominent example certainly is the roughly 10^{10} -fold (!) increase of intracellular ATP/ADP ratios as compared to the equilibrium value in aqueous solution [11,39,40]. Coupling this immense disequilibrium *via* the conditioned hydrolysis of ATP to ADP to the vast majority of (by themselves strongly endergonic) metabolic processes flips the latter's energetics and renders them exergonic. Once a sufficiently high ATP/ADP disequilibrium is attained in a cell, the lion's share of biochemical pathways flows energetically downhill [3,11,38] according to formula [3]. Selected reactions may also be driven by ADP/AMP or related disequilibria and in a number of cases it has been shown that even pyrophosphate/phosphate (PP_i/P_i) disequilibria (sometimes involving higher polyphosphates) can drive biochemical pathways [41-45].



As summarized in Fig. 2, biological free energy conversion therefore can be cooked down to the transformation of environmental redox tensions into the two intracellular chemical disequilibria detailed above. First the environmental redox disequilibrium is converted into a cation-motive gradient between the two sides of the dielectric barrier encapsulating the "living" entity. This cation-motive gradient is first and foremost an electric field but may also (depending on the buffering capacity of the aqueous medium on either side of the barrier) contain an osmotic contribution in the form of a pH-gradient. A few selected species replace the proton- by a sodium gradient in which case both electrostatic and osmotic components necessarily contribute to the cation-motive disequilibrium [46] since sodium ions cannot be buffered away by protonatable groups. This cation-motive gradient is then, in a second step, alleviated (a) *via* machineries embedded within the dielectric barrier through mechanisms coupling the downhill, exergonic flow of cations through the barrier to the condensation of phosphate-anions [39] (see Fig. 2) and (b) in certain species *via* proton-coupled electron transfer processes generating highly reducing redox compounds ("reverse transfer", discussed in section 2.3.1.2.).

It is noteworthy that this 2-step free energy transformation isn't mere catalysis! Catalysis facilitates (speeds up) exergonic reactions while the generation of intracellular disequilibria involves free energy transforming "engines" [3,11] the general principle of which can indeed be likened to that of the steam engine.

2.3. How does bioenergetic conversion of disequilibria actually work?

2.3.1. Up-conversion of reducing power

The generation of reduced ferredoxin and/or NAD(P)H in extant life is achieved by a few functionally dissimilar mechanisms, the only common denominator of which (except for the first one detailed below: photosynthesis) is their fundamentally electrochemical character.

2.3.1.1. Photochemistry-derived reducing power

The light-induced charge separation of an electron/hole pair in photon-excited chlorophyll or bacteriochlorophyll molecules into a pigment-cation and an electron flowing down a chain of acceptor redox centres yields reducing equivalents at very low redox potentials, *i.e.* able to directly (exergonically) reduce ferredoxins. This process certainly beats by far all other types of redox-boosting schemes in terms of absolute occurrence in the biosphere since it is performed by all oxygenic phototrophs (plants, algae and cyanobacteria) and by a portion of the anoxygenic phototrophs. However the harvesting of free energy from sunlight probably did not exist in earliest life [35] and therefore is irrelevant for the quest of tracing back the characteristics of present life towards its emergence.

2.3.1.2. Boosting reducing power through membrane potential

A mechanism frequently employed by species not limited by environmental sources of free energy (*i.e.* many anoxygenic phototrophs as well as some non-phototrophs in environments rich in redox free energy) consists in using a fraction of the cation-motive potential to drive electrons seemingly uphill onto low potential acceptors (see Fig. 3). The vast majority of electron flow from reductants to oxidants (thick arrow in Fig. 3) builds up cation-motive potential as detailed below (section 2.3.2.) while a small fraction (indicated by the thin arrow) gains reducing power *via* partial re-collapsing of this gradient. In the field of bioenergetics this mechanism is termed “reverse electron transfer” [39].

2.3.1.3. The disproportionation-like redox cascade in methane oxidation

Methanotrophic bacteria oxidize methane to methanol and subsequently through formaldehyde and two C1-bodies to eventually produce CO₂. In each of these subsequent oxidation steps, the released electrons are increasingly reducing (Fig. 4). The three terminal redox reactions produce reducing equivalents able to regenerate NADH from NAD⁺. We emphasize that the biological side of these reactions (*i.e.* the involved enzymes) is not responsible for the increase of reducing power which is solely determined by the electrochemical properties of the respective C1-bodies. The intrinsically electrochemical character of this redox-disequilibrium generating reaction scheme has prompted us to count methane-oxidation among the potentially most ancestral energy converting

processes [47].

2.3.1.4. *Electron bifurcation*

In the field of bioenergetics, the term “electron bifurcation” denotes a process wherein the two individual electrons resulting from the oxidation of a 2-electron redox compound (*e.g.* substituted quinones, flavins and potentially molybdo/tungsto-pterins) proceed through thermodynamically divergent routes [48]. While one electron goes on to reduce in a thermodynamically well-behaved manner an electron acceptor with a standard redox potential more positive than that of the 2-electron reductant, the second electron appears to perform an endergonic redox reaction as illustrated in Fig. 5a. The characterized enzymes performing electron bifurcation appear to almost perfectly couple the exergonic and endergonic redox processes and feature extraordinarily low levels of futile side-reactions [49]. The molecular mechanisms underlying this reliable coupling were hotly debated over several decades in the bioenergetics community. The present rationalization is based on an empirically observed [50-53] highly cooperative character of the 2-electron oxidation of the donor molecule resulting in strongly inverted redox potentials for the individual redox transitions as indicated in Fig. 5b. As is obvious from this scheme, the first electron transfer event from D to A_{highpot} is highly endergonic and its completion is therefore contingent on/conditioned by the occurrence of the second one pulling the combined redox reaction into the exergonic regime.

Yuly et al. [54] recently demonstrated *via* the simulation of electron transfer dynamics that the strength of coupling in electron bifurcation is a function of the degree of endergonicity of the first oxidation event in line with the original hypothesis postulating an “escapement”-type entanglement of the two individual redox transitions [51]. The ensemble of our present understanding of electron bifurcation therefore indicates that this mechanism is likely based on purely electrochemical principles and may possibly even occur in abiotic settings once the crucial requirements with respect to relative redox potentials and spatial arrangement are met.

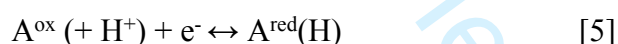
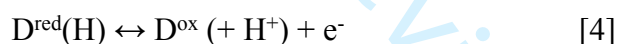
Judging from the end-result of electron bifurcation, it may be regarded as an electrochemical analog to solid state DC-to-DC converters, fairly complex electronic devices [55] routinely used in photovoltaic installations to boost efficiency under dim-light conditions. Compared to these devices, electron bifurcation appears almost ridiculously simple and the goal of emulating this mechanism in abiotic system seems worth pursuing.

While cooperative 2-electron redox transitions remain somewhat mysterious to many biologists, they represent a rather trivial concept in electrochemistry. Obviously, for redox cooperativity to arise, the respective redox compounds need to be in protic solvents. In aprotic solvents (or protic solvents at very high pH), 2-electron compounds such as quinones or flavins feature the intuitive order of individual redox potentials with the first reduction step requiring less

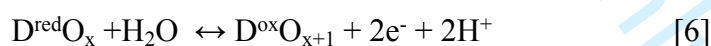
reducing power than the second one, implying thermodynamic stability of the semi-reduced state in between the two redox transitions. Increasing proticity of the solvent lowers the difference in redox potential eventually leading to potential inversion and concomitant thermodynamic instability of the semi-reduced state. This implies that electron bifurcation readily occurs in water but would be precluded in organic solvents such as the oft-cited methane lakes on the Jupiter-moon Titan, invoked by the organic-molecules-centric origin-of-life community as an intriguing environment for life to emerge [56]. By contrast, on water-worlds such as planet Earth, electron bifurcation appears as a very attractive candidate for pre-biotic disequilibrium conversion helping life to come into being [57,58]. We will revisit the question of water's importance for life in section 2.5.

2.3.2. Generation of the cation-motive potential

As emphasized in section 2.1., the ultimate driving redox reaction is an electron transfer from a reduced donor substrate D^{red} to an oxidized acceptor A^{ox} . Depending on the relative values of solution pH and substrate pK, the respective redox transitions are accompanied by protonation/deprotonation events according to



Certain redox substrates furthermore involve oxo-transfers in their redox transitions also entailing the participation of protons:



While electrons appear upon oxidation of D^{red} only to re-disappear in the reduction of A^{ox} , the protons are not compensated if the two individual redox reactions occur on opposite sides of the dielectric barrier. Appropriate localizations of these individual redox processes therefore yield the generation of an ion-motive force which may, for the case of strong buffer capacities on both sides of the barrier, correspond to a simple electric field.

In addition to these directly substrate-linked protonation/deprotonation events, extant life features two further mechanisms participating in build-up of the cation-motive force.

2.3.2.1. Proton shuttling across membranes via quinone redox transitions

In the vast majority of prokaryotic bioenergetic systems, quinone molecules serve as H_2 -

carriers within and across the dielectric barrier *via* their redox transition:



Accordingly, just as for the ultimate redox substrates, the oxidized/reduced transitions of quinones will add to the cation-motive potential if appropriately positioned with respect to barrier-sidedness [9].

2.3.2.2. Proton pumping

Specific bioenergetic enzymes (such as for example Complex I and Complex IV in the mitochondrial electron transfer chain) couple substrate redox transitions to the active pumping of protons through the dielectric barrier [39,59]. In most of these enzyme systems, the mechanistic details are still debated. However, the fundamental functional principle must be based on redox-linked protonation/deprotonation as becomes obvious when the respective processes are stripped down to their bare minimal reaction schemes. Fig. 6 illustrates this fact for the case of mitochondrial Complex I.

2.4. Who is doing the job?

The standard paradigm in biology is that bio-catalysis is performed by enzymes. At least for the case of bioenergetics, a closer look suggests that within these enzymes, it is mainly at the inorganic components, *i.e.* the transition metals and clusters thereof, where the actual action happens. Undeniably, some organic molecules such as NAD, quinones, flavins, pterins, folates and a few more are also crucially involved in the respective processes. Fig. 7 shows a highly stylized rendering of a free energy converting system close to home, the mitochondrial respiratory chain, highlighting the organic/inorganic character of the involved redox centres. Obviously, inorganic metal centres fairly outnumber the organic redox molecules. Similar domination of metal centres of organics is observed in all prokaryotic free energy converting systems we are aware of. Bioenergetics therefore appears to be mainly performed by inorganic entities which led us and other colleagues to propose scenarios for life's emergence wherein a rudimentary form of bioenergetics appeared in the context of certain minerals exposed to environmental electrochemical and pH-gradients [60-64]. For mineralogical and thermodynamics details of these scenarios, we refer the reader to our dedicated articles [3,8,11,32,47,66-69].

2.5. The crucial role of water in life's emergence; one more reason!

In sections 2.1. to 2.3. we have tried to identify the essential physico-chemical parameters of

the free energy converting processes driving life to emerge in the form of a dissipative structure as defined by far-from-equilibrium thermodynamics [32,9,70]. Obviously, all these processes heavily rely on electrochemical principles. More specifically, they are based on aqueous electrochemistry! The coupling of electron transfer to protonations/deprotonations as well as cation transport across the dielectric barrier would be unthinkable in aprotic organic solvents. The same holds true for all the intricate redox reactions involving cooperative 2-electron redox transitions, wherein electron bifurcation is only one specific, albeit extraordinarily important case. Life's free energy conversion is contingent on the possibility that redox state of a cofactor influences pKs of neighbouring protonatable groups and, *vice versa*, that protonation state of a cofactor or its environment modulates the reduction potential of the cofactor. Incidentally this crucial coupling of redox reactions and protonation state is among the numerous reasons why we have proposed the double-layered Fe-oxyhydroxide minerals as potentially pivotal in life's emergence. The details of the elaborated scenarios exceed the scope of this article and we address interested readers to the respective literature [66-69,71,72].

3. Conclusion

As we have tried to convey in this contribution, the most fundamental process of life, that is, biological free energy conversion, is first and foremost accomplished by inorganic entities and relies entirely on the principles of electrochemical reactions in aqueous media. Basically all of the individual reactions are conceivable in fully pre-biotic, inorganic settings. This observation suggests a possible direct link between bioenergetics in extant life and primordial free energy converting processes. The biological free energy converting principle as outlined in this article may thus have been constant since life's very emergence. This should not come as a surprise since free energy conversion is the thermodynamic *sine-qua-non* of life's very existence and fundamental ruptures and *ab initio* innovations replacing the former principle are thus extremely unlikely. The emergence of photosynthesis, often cited as an evident bioenergetic rupture, in fact provides a telling example of life's loyalty to the founding bioenergetic mechanism: While the penultimate sources of free energy shifted from geochemical ones (based on radioactivity-derived heat in Earth's mantle and resulting convection-driven generation of redox disequilibria) to solar photon energy (based on nuclear fusion), the detailed individual mechanisms, that is, the specific enzymes and redox cofactors within, as well as the electron transfer pathways remained basically immutable [9]. Only the source of redox tensions changed when photosynthesis emerged allowing straightforward use of the pre-existing processes.

The bioenergetic principle of extant life therefore seems to be the basic invariable which may provide the Ariadne's thread leading the way out of the maze of scenarios for life's emergence.

Following this thread, we and others [10,36,47,60,63-69,73-75] have in the past put forward tentative proposals for plausible prebiotic settings appealing to specific minerals (fougerite, mackinawite and greigite) within chimneys of alkaline hydrothermal vents. From what was said above, we would venture that progressing in our understanding of life's emergence in such settings will require extensive input of expertise from the community of electrochemists. The question of the emergence of life may to our minds fall much more in the realm of the physical than the life sciences.

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Figure Legends

Figure 1: The universal source of Gibbs free energy in extant life corresponds to an electrochemical tension between reductants and oxidants. In the vast majority of cellular life on Earth, this tension is the result of an environmental disequilibrium in certain redox substrates. In the abscissa on the left, “reducing” and “oxidizing” denote increasingly negative and increasingly positive redox midpoint potentials of the respective redox couples.

Figure 2: Schematic representation of the fundamental mechanism of free energy conversion in life. The environmental redox disequilibrium and the share of free energy (here indicated as flux of enthalpy dH/dt) driving life’s entropy reduction (TdS/dt) are indicated in pink and blue boxes, respectively. F_d stands for ferredoxin. The curved gray bar indicates the electrostatic barrier, i.e. the lipid membrane, with cellular inside to the top and the environment to the bottom of the barrier. Electron transfer is indicated by dashed lines, proton translocations by dotted lines and the substrate redox conversions are denoted by a continuous line. The electrostatic potential resulting from the electron transfer processes and driving proton movement towards the inner compartment is depicted as a green double-headed arrow ($\Delta\psi$).

Figure 3: Schematic representation of the generation of reducing equivalents (eventually NADH) by driven by the electron-transfer-generated cation- (here proton-) motive force. Build-up of the cation-motive force proceeds as indicated in Fig. 2 and a fraction of this potential serves to reverse the redox equilibrium between the reducing substrates and the NADH/NAD⁺ couple *via* coupling to the partial collapse of the ion-motive force. Electron transfer events are denoted by continuous blue arrows while cation- (proton-) transfer is marked by red dashed arrows. Redox conversions are shown by continuous purple lines. In bioenergetics this process is somewhat misleadingly called “reverse electron transfer”.

Figure 4: Redox scheme of the “methanotrophic” oxidation cascade of methane to methanol by molecular oxygen and onwards up to the formate/CO₂ couple. The horizontal bars represent redox midpoint potential with respect to the normal hydrogen electrode (NHE) for the indicated redox couples. Remarkably, these redox midpoint potentials decrease for increasingly oxidized couples to eventually become more reducing than the NADH/NAD⁺ couple (dashed horizontal line).

Figure 5: The mechanism of electron bifurcation. The left scheme (a) shows the experimentally observable events. A 2-electron donor is oxidized and reduces two distinct 1-electron acceptors. One of these acceptors (A_{highpot}) features a redox midpoint potential more positive than the 2-electron donor while that of the second one (A_{lowpot}) is more negative. The total redox reaction is exergonic, that is, the difference between the potentials of the donor and the low potential acceptor is lower than that between the donor and the high potential acceptor. The scheme therefore seems to couple an endergonic half-reaction to an exergonic one. The origin of the coupling was long debated and is now mainly rationalized by cooperative redox transitions for the 2-electron donor resulting in strongly inverted individual potentials. As discussed in the text, this renders the first oxidation step highly endergonic and thus contingent on the occurrence of the second oxidation step. The redox potential abscissa is defined as in Fig. 1.

Figure 6: Proton pumping (a) in a real-life example, Complex I of the mitochondrial respiratory chain and (b) reduced to its electrochemical essentials. The detailed mechanism in Complex I, coupling electron transfer from NADH to quinone is still debated. As suggested in the stripped-down version depicted in (b), from the electrochemical point of view, this mechanism must necessarily involve redox-state-dependent pKs and protonation-state dependent midpoint potentials. The red dashed arrow denotes proton-translocation, the continuous blue one stands for electron transfer and the continuous purple arrows represent changes in redox state.

Figure 7: Stylized representation of the mitochondrial respiratory chain, that is, the free energy converting system of eukaryotes (including humans). Blue arrows denote electron transfer pathways while brown ones stand for movements of the redox cofactor (a quinone) between binding sites on the respective enzymes.

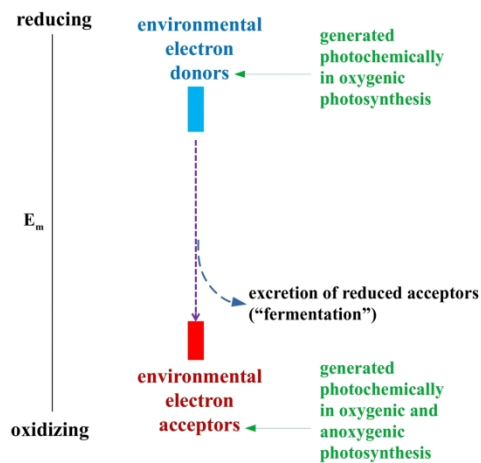


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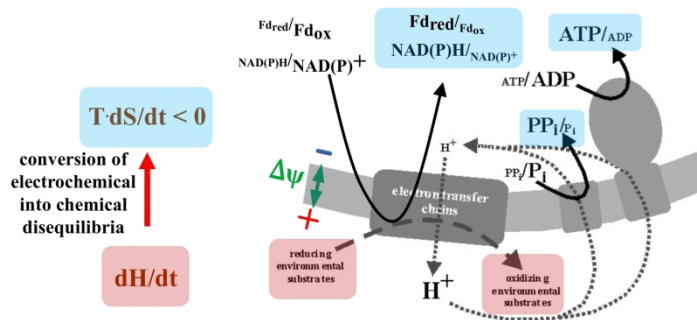


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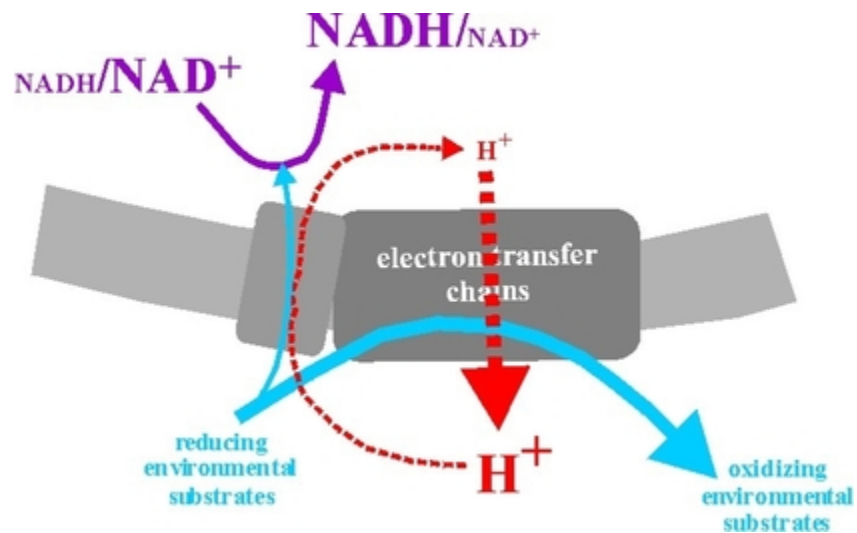


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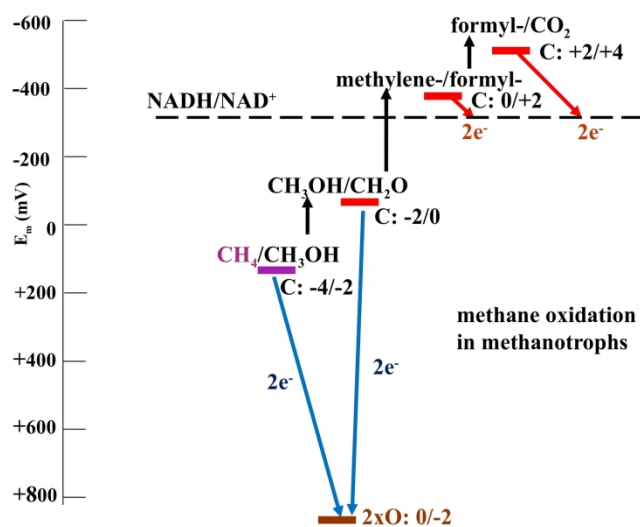


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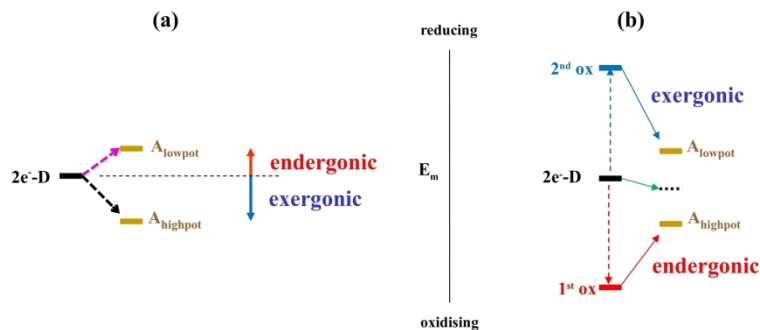


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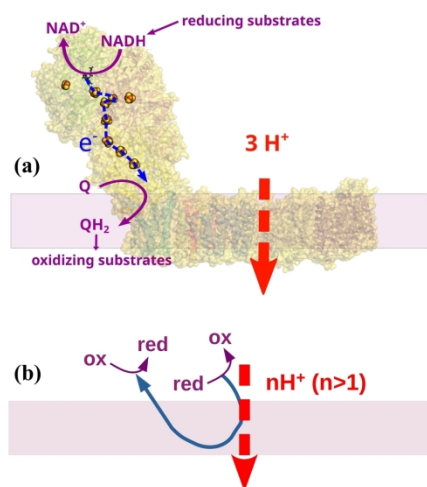


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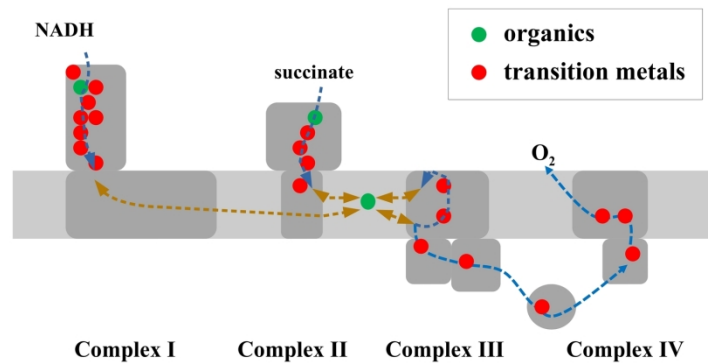


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