

resolution of the ETC complexes in brain mitochondria. The protein complexes of the mitochondrial oxidative phosphorylation system have been reported to form supramolecular assemblies termed respiratory supercomplexes or respirasomes. BN-PAGE was used in this study to analyze the mitochondrial subunit assembly into respiratory chain complexes in rat brain synaptic and non-synaptic mitochondria. Using the mild detergent digitonin for solubilisation of mitochondrial membranes, it was shown that complexes I and II–V interact to form supercomplexes. However, initial experiments suggest that the super-complex composition is different between synaptic and non-synaptic mitochondria from rat brain. The consequences for such disparity in supercomplex formation will be discussed.

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S16.7 Analysis of proteins released through the permeability transition pore of rat brain mitochondria

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Mitochondrial dysfunction can contribute to cell death by not only energetic failure and increased ROS production, but also by the activation of the mitochondrial permeability transition pore (PTP) and release of proapoptotic proteins. The PTP complex is a dynamic poly-protein complex, which spans both mitochondrial membranes at the contact site. An elevation of matrix calcium, beyond a critical threshold, is one of the strongest inducers of the pro-apoptotic PTP. Differing flux control coefficients and energy thresholds have been recorded between synaptic and nonsynaptic mitochondria extracted from rat brain, however, little is known about the proteins that are released from their respective PTPs under stressful conditions. In this study we investigated the calcium-induced swelling in energized/de-energized synaptic and non-synaptic rat brain mitochondria. We report that rat brain PTP opening (as measured by swelling) in both types of mitochondria was more sensitive to Bongkrekic acid than to Cyclosporin A. Furthermore, following swelling of the mitochondria, the proteins released through the pore were resolved on a 2D-PAGE and identified by MALDI-TOF mass spectrometry. The differences between the proteins released through the PTPs from synaptic and non-synaptic rat brain mitochondria and their physiological implications will be discussed.

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(C1) Genomics and evolution colloquium lecture abstracts

C1/1 Introductory notes: Energetic constraints at the very beginning of life

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The number of hypothetical scenarios for the origin of life is unlimited. The space of possibilities, however, can be dramatically restricted by consistently invoking physical, chemical, biological and geological constraints. The short introductory talk will focus on energetic constraints, in particular on the consideration of energy

sources that could be available and utilizable at the earliest stages of evolution.

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C1/2 Energetics of the first bacteria as inferred from genome analysis

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The availability of complete genome sequences had a major impact on modern biology, resulting in a much better understanding of cell metabolism. Owing to their complex subunit structure, membrane energy-transducing complexes remained outside the scope of most comparative-genomic analyses. We compared the distribution of proton- and sodium-translocating enzymes encoded in bacterial and archaeal genomes, analyzed the physico-chemical and evolutionary constraints for their origin, and used these data to infer an evolutionary scenario for the origin of the energy transduction machinery. Surprisingly, results of comparative structural and phylogenetic analyses suggest that sodium-translocating ATP synthases and ion pumps preceded the proton-translocating ATP synthases and proton pumps. Thus, the first prokaryotes likely relied on sodium ion gradient for their energy metabolism. Proton-based energetics must have emerged later, following the development of proton-tight membranes through different adaptations in bacteria, archaea and eukaryotes. Evolutionary advantages of proton-based energetics, in particular, chemical coupling of transmembrane proton translocation with electron transfer from organic substrates to terminal electron acceptors, such as oxygen or nitrate, ensured wide dissemination of the corresponding genes and resulted in the switch from Na⁺ to H⁺ as the coupling ion in most bacteria and archaea. Currently, sodium-based energetics is found primarily in obligate anaerobic prokaryotes, including some important human pathogens.

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C1/3 Chloroplast sensor kinase – The redox messenger of organelle gene expression

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Why are there genes in chloroplasts and mitochondria? The CoRR hypothesis states that organellar genes and their gene products are Co-located for Redox Regulation. CoRR predicts (i) that an irreducible core of genes must be retained by chloroplasts and mitochondria from their bacterial ancestors, and (ii) that a bacterial redox signalling pathway exerts regulatory control over expression of these genes, using components that have operated continuously throughout the transition from prokaryote to bioenergetic organelle. Chloroplast Sensor Kinase (CSK) is a chloroplast stromal protein that is the product of the nuclear gene *At1g67840* of *Arabidopsis thaliana*. T-DNA insertion lines are impaired in plastoquinone redox control of transcription of chloroplast genes for reaction centre apoproteins of photosystem I and II and do not adjust PS I/PS II stoichiometry. CSK is homologous with bacterial histidine sensor kinases and yet is universal in photosynthetic eukaryotes. We propose that CSK provides the redox regulation