transfers of their operons from bacteria; such transfers would depend on the presence of the archaea-specific systems of fatty synthesis in the respective archaeal genomes [4]. The gradual oxygenation of the atmosphere would have driven further evolution since auto-oxidation of the components of the cytochrome bc complex results in the generation of potentially hazardous reactive oxygen species. The choice of different means of decreasing the extent of autooxidation is proposed to have driven the divergence of the b6f-type complexes of cyanobacteria (ancestors of chloroplasts) and the bc1-type complexes of respiring alpha-proteobacteria (ancestors of mitochondria).

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S11.I4

Molecular basis of photoprotection in photosynthetic organisms

Bruno Robert
Institute of Biology and Technology CEA Saclay, 91191 Gif/Yvette Cedex, France
E-mail address: bruno.robert@cea.fr

Through their light-harvesting antenna, plants frequently absorb more solar energy than they can use in photosynthesis. This excess energy has the potential to cause cell damage, such as pigment bleaching and protein inactivation. To minimise photodamage, a number of protection mechanisms exist, which we have characterised at a molecular/functional level. I will discuss the following mechanisms in the light of our most recent results

Protection against singlet oxygen production by chlorophyll triplet states

In most light-harvesting proteins of oxygenic organisms, ultrafast triplet–triplet transfer is associated with a significant alteration of the triplet states involved. Experiments performed with synthetic dyads suggest that such alteration, which is likely to be due to the presence of intramolecular charge transfer states, is necessary for ultrafast triplet–triplet transfer to occur.

Protection against photooxidative stress induces by high light environments

Excitation energy quenchers rapidly appear in the photosynthetic membrane of plants and algae when these organisms are exposed to high illumination conditions. In the last decade, combined use of advanced spectroscopic methods, first applied on isolated light-harvesting complexes then developed for their application on systems as complicated as whole leaves, has yielded a precise picture of the molecular events which underlie this important mechanism of photoprotection in higher plants. Recent results on diatoms suggest that the photoprotection mechanisms are quite similar in these algae, although the structure of their light-harvesting apparatus is quite different.

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S11.O1

Modification of spin coupling between the semiquinone and Rieske cluster in the Qo site of cytochrome bc_1

Artur Osyczka, Łukasz Bujnowicz, Małgorzata Dutka, Sebastian Pintscher, Marcin Sarewicz
Jagiellonian University, Krakow, Poland
E-mail address: artur.osyczka@uj.edu.pl

Usually in electronic bifurcation associated with oxidation of hydroquinone at the Q_o site of cytochrome bc_1 (mitochondrial complex III) awaits description of its molecular mechanism. Recently, we discovered two distinct populations of semiquinone (SQ_o) associated with operation of the Q_o site: i) SQ_o coupled to the reduced Rieske cluster (FeS) via spin–spin exchange interaction and ii) a separate, unusually fast-relaxing SQ_o [1]. Here, using freeze-quench and electron paramagnetic resonance spectroscopy, we examined in detail conditions of formation of these two populations of SQ_o under a variety of buffer and reaction mixture compositions. The analysis included native cytochrome bc_1 and specific mutants that affected the motion of the FeS subunit. The revealed differences in proportions between the SQ_o signal and the SQ_o–FeS coupled signal indicated that these signals are inversely correlated with one another. Shifting FeS toward the Q_o site increases the amount of SQ_o–FeS coupled system at the cost of SQ_o, while shifting the FeS out of the Q_o site has the opposite effect — increase in SQ_o at the cost of SQ_o–FeS coupled signal. We anticipate that these results will contribute to better understanding of the electron reactions that take place within the Q_o site, in particular the formation of SQ_o intermediates and their relation to superoxide generation by the Q_o site.

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S11.O2

A photo-useless chlorophyll: What's for?

Francesca Zito[a], Audrey Le Bas[a], Silvia Zambolin[a], Agnes de Lacroix de Lavalette[a], Lise Barucq[a], David Stroebel[a], Yves Pierre[a], Fabrice Rappaport[b], Daniel Picot[a]
[a]*Laboratoire de Biologie Physico Chimique des Protéines Membranaires, CNRS, Paris Diderot UMR 7099, Italy*
[b]*Laboratoire de Physiologie Membranaire et Moléculaire du Chloroplaste, CNRS, UPMC UMR 7141, Italy*
E-mail address: zito@ibpc.fr

3D structure of the membrane complex cytochrome b6f from the green alga *Chlamydomonas reinhardtii* was solved in the presence and in the absence of a Qo site inhibitor (TDS) (1). The analyses of the two structures revealed, beside the reorganisation of the Rieske soluble domain, small conformational changes around the Qo site. Cytochromes b6f and bc1 are structurally and functionally very similar but they have a different sensitivity to inhibitors and in the cytochrome b6f the phytil chain of a chlorophyll molecule stands at the entrance of the Qo site. The presence of this molecule raises the question of its role which is still unknown. In order to gain information on chlorophyll function, its binding to the b6f complex was destabilised by site-directed mutagenesis of the petD gene. Alanine 140 in helix G was replaced by phenylalanine, a bulkier amino acid, this position faces the pigment, but remains away from

the Qo site. We show that removal of chlorophyll does not impair the overall structure of the b6f complex, but perturbs in vivo and in vitro plastoquinol oxidation in the Qo site, disproving a simple structural role for the chlorophyll. In addition, this compromised activity can be correlated with conformational changes, which comprises the same polypeptide segments that are also subject to conformational changes in the related Qo sites of the bc1 and b6f complexes upon inhibitor binding. This common flexibility might be involved in the gating of the Qo site to avoid short circuits, introducing a double gating.

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S11.P1

Lutein as a mediator of effective yet 'economic' regulator of light-harvesting in photosystem II of plants

Christopher Duffy[a], Erica Belgio[a], Jevgenij Chmeliov[b], Leonas Valkunas[b], Alexander Ruban[a]
[a]*Queen Mary University of London, United Kingdom*
[b]*University of Vilnius, Lithuania*
E-mail address: c.duffy@qmul.ac.uk

The antenna of photosystem II (PSII) in plants possesses an intrinsic photoprotective switch that allows for rapid adaptation to changing light intensity. The essence of this switch is the formation of energy-quenching sites within the antenna that trap and dissipate excess excitation energy, thereby mitigating the photoinhibitory damage associated with high light [1]. The molecular mechanisms at the heart of this process, known as Non-Photochemical Quenching, are still debated and there are several competing models [1]. One such model, based on time-resolved fluorescence measurements, suggests that NPQ occurs via the incoherent transfer of energy from chlorophyll to the xanthophyll lutein 1, followed by rapid (~10 ps) non-radiative decay, in the major PSII antenna complex LHCII [2]. Our recent theoretical model of chlorophyll–xanthophyll energy transfer in LHCII indicates that this mechanism is sufficient to explain the fluorescence quenching in LHCII crystals [3]. Additionally we reported that a combination of solid-state NMR and theoretical modelling implies that aggregation-induced quenching in LHCII may be controlled by fine tuning of chlorophyll–xanthophyll distances [4]. We here present the incorporation of this lutein-mediated quenching mechanism into coarse-grained models of energy transfer in PSII-antenna superstructures and show that lutein is an effective NPQ site in the photosynthetic membrane. Additionally, we show that this model is consistent with our recent fluorescence induction measurements [5]. We show that lutein in the PSII antenna can function as an 'economic' quencher, offering protection to closed reaction centres whilst simultaneously preserving photosynthetic productivity in a broad range of light intensities.

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P S11.P2

Inter-monomer electron transfer in asymmetrically mutated fusion hybrid cytochrome bc1-like complex supports the enzyme function in vivo

Robert Ekiert, Marcin Sarewicz, Artur Osyczka
Jagiellonian University, Krakow, Poland
E-mail address: robert.ekiert@uj.edu.pl

Cytochrome bc_1 is one of the key catalytic complexes involved in the bioenergetic processes. This dimeric enzyme operates according to the Q cycle functionally linking quinone catalytic sites on the opposite sides of the membrane. While recent studies indicate that this link can be achieved by intra- or inter-monomer electron transfer, the physiological relevance of the latter remains unclear. To address this issue we used a system based on a fusion hybrid protein build of cytochrome *b* subunits from two different species: *Rhodobacter capsulatus* and *Rhodobacter sphaeroides*. It assembles into a functional bc_1-like complex and allows introduction of mutations in an asymmetric manner [1]. We report that the cross-inactivated bc_1 constructs, enforcing the inter-monomer transfer as the only way to electronically connect the opposite catalytic sites, supported the cytochrome bc_1-dependent photoheterotrophic growth of bacteria. This indicates that inter-monomer route for electrons is able to sustain the function of the enzyme in vivo. Moreover, given the differences in amino acid composition between the fused cytochromes *b*, we suggest that the minimal requirement for the enzyme bioenergetic efficiency is to provide the uninterrupted electron connection of the quinone catalytic sites on the opposite sides of the membrane in a manner robustly tolerant to structural alterations.

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S11.P3

Role of electrostatic and hydrophobic interactions in the encounter complex formation of plastocyanin and cytochrome f

Johannes M. Foerster[a], Sandra Scanu[b], G. Matthias Ullmann[c], Marcellus Ubbink[b]
[a]*Computational Biochemistry/Bioinformatics*
[b]*Institute of Chemistry, Leiden University, The Netherlands*
[c]*Computational Biochemistry, University of Bayreuth, Germany*
E-mail address: johannes.foerster@uni-bayreuth.de

Cytochrome f (cyt f) and plastocyanin (pc) are electron transfer proteins forming a transient complex. During complex formation, an initial encounter complex rearranges into an active complex. Both, electrostatic and non-covalent short-range forces have been reported to be important for the complex in *Phormidium laminosum* or Nostoc [1]. In this work, the association of cyt f and pc were studied using paramagnetic NMR spectroscopy, Monte Carlo simulations [2] and ensemble docking in order to get deeper insights into the dynamics of the electron transfer complexes. For this purpose, spin labels were attached to cyt f, and relaxation enhancements of pc nuclei were measured, demonstrating that a large part of the cyt f surface area is sampled by pc. The distribution of the encounter complex