

Regulation and Dynamics of the Light-Harvesting System

Jean-David Rochaix

Departments of Molecular Biology and Plant Biology, University of Geneva, 1211 Geneva, Switzerland; email: jean-david.rochaix@unige.ch

Annu. Rev. Plant Biol. 2014. 65:287–309

First published online as a Review in Advance on January 22, 2014

The *Annual Review of Plant Biology* is online at plant.annualreviews.org

This article's doi:
10.1146/annurev-arplant-050213-040226

Copyright © 2014 by Annual Reviews.
All rights reserved

Keywords

photosynthesis, thylakoid membrane, light acclimation, state transitions, nonphotochemical quenching

Abstract

Photosynthetic organisms are continuously subjected to changes in light quantity and quality, and must adjust their photosynthetic machinery so that it maintains optimal performance under limiting light and minimizes photodamage under excess light. To achieve this goal, these organisms use two main strategies in which light-harvesting complex II (LHCII), the light-harvesting system of photosystem II (PSII), plays a key role both for the collection of light energy and for photoprotection. The first is energy-dependent nonphotochemical quenching, whereby the high-light-induced proton gradient across the thylakoid membrane triggers a process in which excess excitation energy is harmlessly dissipated as heat. The second involves a redistribution of the mobile LHCII between the two photosystems in response to changes in the redox poise of the electron transport chain sensed through a signaling chain. These two processes strongly diminish the production of damaging reactive oxygen species, but photodamage of PSII is unavoidable, and it is repaired efficiently.

Contents

| | |
|--|-----|
| INTRODUCTION..... | 288 |
| ACCLIMATION OF THE PHOTOSYNTHETIC MACHINERY | |
| TO CHANGES IN REDOX POISE | 292 |
| ROLE OF THE STT7/STN7 KINASE AND PPH1/TAP38 PHOSPHATASE IN | |
| STATE TRANSITIONS..... | 294 |
| REGULATION OF LIGHT-HARVESTING COMPLEX GENE EXPRESSION | |
| AND ASSEMBLY | 295 |
| ENERGY-DEPENDENT NONPHOTOCHEMICAL QUENCHING | 297 |
| PHOTOSYSTEM II PROTEIN PHOSPHORYLATION AND THYLAKOID | |
| MEMBRANE FOLDING..... | 299 |
| PERSPECTIVES | 301 |

INTRODUCTION

The primary reactions of oxygenic photosynthesis are mediated by four protein complexes embedded in the thylakoid membrane. Three of these complexes—photosystem II (PSII), the cytochrome *b₆f* complex (Cyt*b₆f*), and photosystem I (PSI)—contain chlorophylls, carotenoids, and lipids and are electrically connected in series through the electron transport chain (**Figure 1**). PSII and PSI are associated with their light-harvesting complexes (LHCII and LHCI, respectively). These antenna systems absorb light and channel the light excitation energy to the reaction centers of PSII and PSI, where a stable charge separation occurs across the thylakoid membrane. In this way a strong oxidant is generated on the donor side of PSII that is capable of extracting electrons from water, which are then transferred from PSII along the electron transport chain to the plastoquinone (PQ) pool and Cyt*b₆f* and subsequently to plastocyanin and PSI, where a second photochemical reaction occurs that results in the oxidation of plastocyanin and the reduction of ferredoxin and finally of NADP⁺. In addition to linear electron flow (LEF) from PSII to PSI, cyclic electron flow (CEF) occurs in which electrons are shuttled back to the PQ pool from the acceptor side of PSI. Both LEF and CEF are coupled to the transfer of protons from the stromal to the lumenal side of the thylakoid membrane. The resulting proton gradient is used by the fourth complex, ATP synthase, to produce ATP, which together with NADPH fuels the reactions of carbon assimilation and cellular metabolism.

A characteristic feature of the thylakoid membrane is that it is organized in two distinct functional domains. In most cases, the grana, which are cylindrical stacks of 5–20 layers of thylakoid membrane, are linked together by stroma lamellae, although this may vary depending on plant species and growth conditions. Each granum is bounded by two stroma-exposed membranes and the highly curved margins that connect two adjacent grana membranes. Within each chloroplast, these thylakoid membranes form a continuous network with a single lumen space (116). There is a remarkable lateral heterogeneity in these membranes, as the photosynthetic complexes are not uniformly distributed between the two thylakoid domains (3). PSII is localized mainly in the grana, whereas PSI and ATP synthase are localized in the stroma lamellae and Cyt*b₆f* partitions equally between both domains (**Figure 1**). The heterogeneity of the thylakoid system is determined largely by the fact that both PSI and ATP synthase contain large protruding stroma domains that would not fit within the appressed grana membranes, which are separated by only 3–4 nm (34, 70). Moreover, grana formation is mediated through van der Waals attractive forces

PSII: photosystem II

Cyt*b₆f*: cytochrome *b₆f* complex

PSI: photosystem I

LHC: light-harvesting complex

PQ: plastoquinone

LEF: linear electron flow

CEF: cyclic electron flow

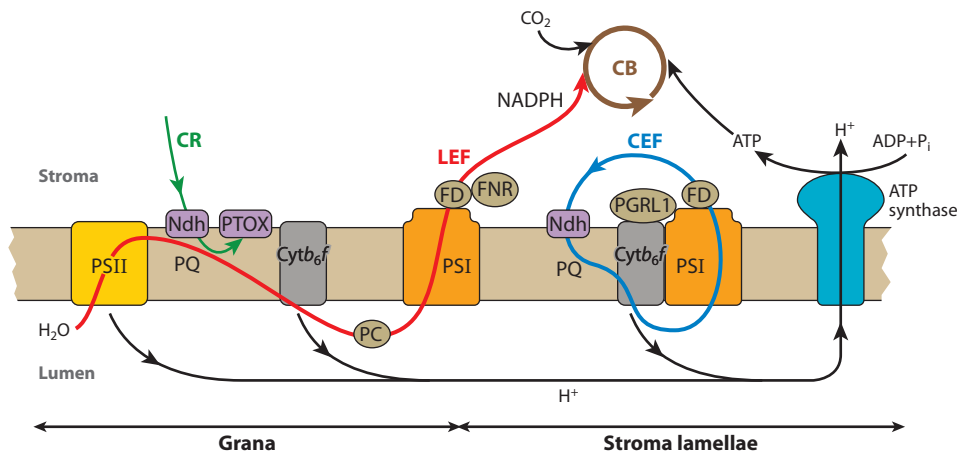


Figure 1

Lateral heterogeneity of the thylakoid membrane with photosynthetic electron pathways, showing the photosynthetic electron transport chain with photosystem II (PSII), cytochrome b_6f (Cyt b_6f), photosystem I (PSI), and ATP synthase. Linear electron flow (LEF) and cyclic electron flow (CEF) are shown in red and blue, respectively, with arrows indicating the direction of electron flow. The LEF pathway is driven by the two photochemical reactions of PSII and PSI. Electrons are extracted by PSII from water and transferred subsequently to the plastoquinone (PQ) pool, Cyt b_6f , plastocyanin (PC), PSI, and ferredoxin (FD). Ferredoxin-NADPH reductase (FNR) catalyzes the formation of NADPH at the expense of reduced FD. The CEF pathway is driven by PSI in the stroma lamellae. In *Chlamydomonas reinhardtii*, PSI forms a supercomplex with Cyt b_6f , FNR, PGRL1, and additional factors. Upon reduction of FD, electrons are returned to the PQ pool through either the NADH dehydrogenase complex (Ndh) or PGRL1, which acts as an FD-PQ oxidoreductase. Both LEF and CEF are associated with proton pumping into the lumen. The resulting proton gradient is used by ATP synthase to produce ATP, which together with NADPH drives CO₂ assimilation by the Calvin-Benson cycle (CB; brown). The chlororespiratory chain (CR; green) feeds stromal-reducing power into the PQ pool with plastid terminal oxidase (PTOX) as the terminal electron acceptor.

and cation-mediated electrostatic interactions between membranes, in which LHCII plays a major role (10, 28).

The LHCII and LHCI proteins are encoded by a large family of genes. The green alga *Chlamydomonas reinhardtii* has 9 major and 3 minor LHCII-encoding genes and 9 LHCI-encoding genes (92), and in the land plant *Arabidopsis thaliana*, 15 LHCII-encoding genes and 6 LHCI-encoding genes have been identified (61) (Table 1). Each LHC protein contains three transmembrane domains with up to eight chlorophyll *a* molecules, six chlorophyll *b* molecules, and four xanthophyll molecules (86). The major LHCII proteins are organized in trimers around the PSII core, which consists of the D1 and D2 protein heterodimer, the chlorophyll *a*-containing core antenna proteins CP43 and CP47, and a set of low-molecular-weight polypeptides. The LHCII trimers are connected to the PSII core through two less abundant monomeric LHCII proteins, CP26 and CP29 (Figure 2). Imaging by transmission electron microscopy revealed PSII-LHCII supercomplexes with different antenna sizes. Land plants contain C₂S₂M₁- and C₂S₂M₂-type supercomplexes (20, 27). (C, S, and M refer to the core complex and strongly and moderately associated LHCII trimers, respectively.) Supercomplexes with one to three LHCII trimers per monomeric core were detected in *C. reinhardtii* (37, 100, 122). Plants also contain another monomeric LHCII protein, CP24, that is required for binding M trimers to the core and is missing in green algae (120).

The PSI complex is monomeric in eukaryotes and consists of the PsaA/PsaB heterodimer and additional surrounding subunits. Most of the chlorophylls bound to the reaction center subunits

Table 1 Light-harvesting complex (LHC) proteins in *Cblamydomonas reinhardtii* and *Arabidopsis thaliana*

| Protein | Number of amino acids ^a | Accession number | Protein | Number of amino acids ^a | Accession number |
|--------------------------------------|------------------------------------|------------------|-----------------------------------|------------------------------------|------------------|
| LHCII, <i>C. reinhardtii</i> | | | LHCII, <i>A. thaliana</i> | | |
| Lhcbm1 | 256 | BAB64418 | Lhcb1.1, -1.2, -1.3 | 267 | X03907 |
| Lhcbm2 | 249 | BAB64417 | Lhcb1.4 | 267 | X64459 |
| Lhcbm3 | 257 | BAB64416 | Lhcb1.5 | 266 | X64460 |
| Lhcbm4 | 254 | AAD03731 | Lhcb2.1, -2.2, -2.3 | 265 | AF134122 |
| Lhcbm5 | 268 | AAD03732 | Lhcb2.4 | 266 | AF134125 |
| Lhcbm6 | 253 | AAM18056 | Lhcb3 | 265 | AF134126 |
| Lhcbm7 | 249 | AAK01125 | Lhcb4.1 (CP29) | 290 | X71878 |
| Lhcbm8 | 254 | AAL88457 | Lhcb4.2 (CP29) | 288 | AF134127 |
| Lhcbm9 | 254 | EDP01724 | Lhcb4.3 (CP29) | 276 | AF134128 |
| Lhcb4 (CP29) | 280 | BAB64415 | Lhcb5 (CP26) | 280 | AF134129 |
| Lhcb5 (CP26) | 289 | BAB20613 | Lhcb6 (CP24) | 288 | AF134130 |
| Lhcb7 | 399 | EDP07628 | | | |
| LHCI, <i>C. reinhardtii</i> | | | LHCI, <i>A. thaliana</i> | | |
| Lhca1 | 243 | BAD06923 | Lhca1 | 241 | M85150 |
| Lhca2 | 257 | EDP05477 | Lhca2.1 | 271 | AF134120 |
| Lhca3 | 228 | BAD06919 | Lhca3 | 273 | U01103 |
| Lhca4 | 241 | BAD06918 | Lhca4 | 252 | M63931 |
| Lhca5 | 264 | BAD06920 | Lhca5 | 255 | AF134121 |
| Lhca6 | 257 | BAD06922 | Lhca6 | 277 | U03395 |
| Lhca7 | 241 | BAD06924 | | | |
| Lhca8 | 267 | BAD06921 | | | |
| Lhca9 | 213 | EDP04026 | | | |
| qE-NPQ, <i>C. reinhardtii</i> | | | qE-NPQ, <i>A. thaliana</i> | | |
| LHCSR1 | 253 | CAA64632 | PsbS ^b | 265 | AF134131 |
| LHCSR3.1 | 259 | EDP01013 | | | |
| LHCSR3.2 | 259 | EDP01087 | | | |
| PsbS ^b | 245 | EDP09214 | | | |

Abbreviation: qE-NPQ, energy-dependent nonphotochemical quenching.

^aCorresponds to the precursor protein, including the transit peptide.

^bIn contrast to the other LHC proteins, which contain three transmembrane domains, PsbS contains four transmembrane domains and does not bind pigments.

act as light-harvesting pigments. Several LHCI proteins are bound asymmetrically as a crescent-shaped belt to the PSI core in both algae and land plants (2, 65).

Photosynthetic organisms are constantly subjected to a changing environment in both the long term (because of seasonal and diurnal changes) and the short term (because of fluctuations in cloud cover, canopy, and wind-induced plant movements). These changes lead to considerable shifts in the intensity and spectral quality of the incident sunlight. Moreover, changes in the availability of CO₂, water, and nutrients and variations in temperature can greatly affect the cellular need for ATP, NADPH, and the products of carbon fixation. Thus, not only excess light but also other environmental changes need to be accommodated. Adjusting the operation of the photosynthetic machinery accordingly is a great challenge for algae and plants.

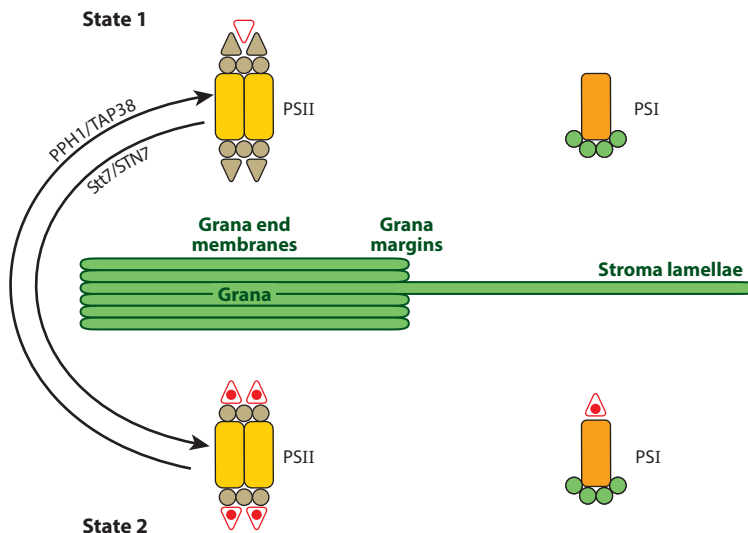


Figure 2

Remodeling of the light-harvesting systems during state transitions. Photosystem II (PSII) is depicted as a $C_2S_2M_2$ complex with monomeric CP29, CP26, and CP24 (circles) connecting the core complex with the light-harvesting complex II (LHCII) trimers (triangles). The transition from state 1 to state 2 is triggered when the plastoquinone (PQ) pool is reduced. Docking of plastoquinol (PQH_2) to cytochrome *b₆f* leads to activation of the Stt7/STN7 kinase and phosphorylation of LHCII (red dots). Phosphorylated LHCII dissociates from PSII and binds to photosystem I (PSI). Upon oxidation of the PQ pool, the kinase is inactivated, LHCII is dephosphorylated by the PPH1/TAP38 phosphatase, and the mobile LHCII returns to PSII. PSII is located in the grana, whereas PSI is located mainly in the stroma lamellae, the grana end membranes, and the grana margins.

A key player in these adaptive processes is the LHCII photosynthetic antenna system, which is involved both in light harvesting and in the protection of the photosynthetic apparatus. Its principal roles are to efficiently collect light energy to drive the primary photochemical reactions of PSII and PSI and to provide photoprotection. To achieve this goal, photosynthetic organisms have developed two distinct strategies. The first regulates the amount of light energy used by the PSII reaction center for photochemistry, especially when this energy exceeds the capacity of the photosynthetic machinery. In this strategy, the system switches to a protective mode in which the excess absorbed energy is dissipated as heat through a process called energy-dependent nonphotochemical quenching (qE-NPQ) (57). Without this protection system, the long-living singlet chlorophyll excited states produced under excess light would undergo triplet formation and react with molecular oxygen to produce reactive oxygen species, which damage cellular components. The second strategy balances the excitation energy absorbed by the LHCII and LHCI antenna systems and readjusts the redox poise of the electron transfer chain upon strong fluctuations in irradiance to optimize photosynthetic electron flow. This readjustment of the absorption cross sections of PSII and PSI can occur in either the short term (through the redistribution of the mobile LHCII antenna, referred to as a state transition) or the long term (through transcriptional and translational regulation of LHC gene expression). In the case of qE-NPQ, the LHCII system is able to sense the state of the light-energy balance directly through the ΔpH across the thylakoid membrane and to respond through structural changes. In the case of state transitions, the system senses changes in the redox state of the PQ pool through a signaling network comprising

qE-NPQ:
energy-dependent
nonphotochemical
quenching

PTOX: plastid
terminal oxidase

Cyt b_6f , protein kinases, and phosphatases, which ultimately restores the redox poise of the electron transport chain and thereby maintains photosynthetic efficiency.

The aim of this article is to cover recent advances in the area of LHCII regulation and thylakoid membrane dynamics. Earlier work has been covered in several recent reviews (55, 69, 79, 111). Because of space limitations, only specific aspects of these topics are discussed here.

ACCLIMATION OF THE PHOTOSYNTHETIC MACHINERY TO CHANGES IN REDOX POISE

The light-harvesting systems of PSII and PSI have a distinct pigment composition. LHCII is enriched in chlorophyll *b*, whereas the antenna of PSI is rich in chlorophyll *a* and additionally contains chlorophylls able to absorb light in the far-red region (13, 32, 78). Changes in light quality and quantity can therefore unbalance the distribution of the absorbed light excitation energy between PSII and PSI. The balance of the system can, however, be restored through state transitions, in which the antenna cross sections of the PSII and PSI antennae are readjusted (22, 97).

Figure 2 shows the classical model of state transitions. Upon overexcitation of PSII relative to PSI, the PQ pool is reduced. This redox state is sensed by a protein kinase, presumably Stt7/STN7 (14, 36), that is activated upon binding of plastoquinol (PQH $_2$) to the Q $_o$ site of Cyt b_6f (124, 134) and phosphorylates a portion of the LHCII antenna (16). Phosphorylated LHCII dissociates from PSII and binds to PSI, thereby rebalancing the excitation energy between PSII and PSI and restoring the redox poise of the PQ pool. This state corresponds to state 2. The process is reversible, as oxidation of the PQ pool by light preferentially absorbed by PSI leads to inactivation of the kinase, dephosphorylation of LHCII by the PPH1/TAP38 phosphatase (108, 114), and return of the mobile LHCII to PSII. This state corresponds to state 1. However, this model is rather simplistic, and recent evidence indicates that the mechanisms of state transitions are far more complex.

Both *C. reinhardtii* and *A. thaliana* have been used extensively to study state transitions and more generally as model systems for green algae and land plants. In this respect, it is interesting to compare acclimation responses in a motile unicellular alga and a sessile multicellular plant, especially with regard to their evolutionary aspects. A major difference between these two organisms is that the mobile LHCII antenna constitutes 80% of the total PSII antenna in *Chlamydomonas* but only 15–20% in *Arabidopsis* (35). In *Chlamydomonas*, the principal function of state transitions appears to be to adjust the ATP level to cellular demands (26, 130). Under conditions leading to a shortage of cellular ATP, such as anaerobiosis, glycolysis is stimulated through the Pasteur effect, and the resulting reducing equivalents are channeled to the PQ pool via the NADH dehydrogenase complex (Ndh in **Figure 1**). The LHCII kinase is then activated, and transition to state 2 occurs. The difference between algae and plants may be only quantitative, as in both cases, the redox state of the PQ pool is influenced by both the metabolic state and the environmental conditions. In both algae and plants, one important role for state transitions is to maintain the redox poise of the PQ pool and respond to metabolic needs (41, 56).

Earlier experiments indicated that transition from state 1 to state 2 in *Chlamydomonas* acts as a switch from LEF to CEF (42, 43). However, recent work has revealed that CEF is controlled by the redox power and is independent of state transitions in *Chlamydomonas* and probably also in land plants (119). The key element was the use of a *Chlamydomonas* mutant deficient in plastid terminal oxidase 2 (Ptox2), which reoxidizes the PQH $_2$ pool in the dark (**Figure 1**). In the absence of Ptox2, this pool remains reduced in the dark even under aerobic conditions and the cells are in state 2, with the mobile LHCII associated with PSI; in the wild type, in contrast, LHCII remains associated with PSII. Under these conditions, however, CEF was not enhanced in the *ptox2* mutant compared with the wild type, indicating that state 2 is not sufficient to promote CEF. Further,

state 2 is not required for inducing CEF, as anoxic conditions enhanced CEF both in the wild type and in an *stt7* mutant blocked in state 1 (119). Also, the PSI-Cyt b_6f supercomplex involved in CEF was formed independently of state transitions (119). These results thus clearly indicate that the efficiency of CEF cannot be correlated with state transitions and that the enhancement of CEF is likely controlled by the redox state of the photosynthetic electron transport chain. How this control operates at the molecular level remains to be determined.

It has become apparent that CEF is important in C₃ plants to match the ATP:NADPH ratio of 3:2 required for driving the Calvin-Benson cycle, particularly under fluctuating light or other stress conditions (64). In the CEF pathway, electrons are transferred back to the PQ pool through NAD(P)H dehydrogenase or through Cyt b_6f using a mechanism derived from the Q cycle (64, 115). Moreover, direct reduction of the PQ pool may occur through ferredoxin-PQ reductase (29, 96). A strong candidate for this reductase is PGRL1 (53), a protein known to be involved in CEF and associated with PSI (33). In *Chlamydomonas*, a large supercomplex that includes PSI, Cyt b_6f , ferredoxin-NADPH reductase, PGRL1, and several additional factors is involved in CEF (59).

Three forms of PSII complexes have been identified in *Chlamydomonas*: a PSII core complex, a PSII-LHCII supercomplex, and a PSII megacomplex containing multiple LHCII (58). The PSII megacomplex is formed in state 1, whereas the PSII core complex is predominant in state 2, indicating that LHCII are dissociated from PSII upon transition to state 2. Also, phosphorylated LHCII is associated mainly with the PSII supercomplex and not with the megacomplex. Based on these studies, PSII remodeling during a state transition was proposed to occur through phosphorylation of LHCII, which converts the megacomplex into a supercomplex, and through the release of LHCII from the PSII supercomplex upon phosphorylation of the minor LHCII CP29 and CP26 and the PSII core proteins (58).

The composition and organization of PSII during state transitions has also been studied in land plants. A PSII supercomplex was identified with four LHCII trimers (C₂S₂M₂). This complex contains up to two additional loosely associated trimers in state 1 (126). During transition to state 2, this extra LHCII migrates to PSI, leaving the PSII supercomplex intact (**Figure 2**). Earlier in vitro studies based on subfractionation of thylakoid membranes indicated that stroma lamellae enriched in PSI contain more phosphorylated LHCII than the grana fraction, which contains predominantly PSII (12, 77). However, in contrast to the classic model, phosphorylation is not restricted to the mobile LHCII but also occurs for the LHCII in the supercomplexes, indicating that the PSII supercomplex does not change during state transitions and that PSII phosphorylation is not sufficient to dissociate certain LHCII trimers from PSII (126). It was assumed earlier that LHCII are the major antenna of PSII and that, in response to exposure to low light, a small fraction of LHCII is displaced to PSI. However, it now appears that a large part of the PSI complexes binds one LHCII trimer, which transfers excitation energy efficiently to PSI after long-term acclimation to different light conditions (low light, moderate light, high light, sunlight) (126, 127). Thus, LHCII is part of the PSI antenna under a variety of growth conditions. Calculations of absorption balance indicate that if all LHCII were associated with PSII, it would be overexcited at wavelengths below 681 nm, but in reality a balance between PSII and PSI occurs when the PSI antenna is enlarged by LHCII.

The picture that emerges from these studies is that, in land plants, the mobile LHCII acts as a highly efficient antenna for PSI under a variety of light conditions and remains associated with PSI after long-term acclimation. The mobile LHCII moves back to PSII only under specific conditions, such as after a sudden increase in light intensity or when PSI is overexcited by far-red light. In mutants lacking the PsaH subunit that is essential for docking LHCII to PSI, phosphorylated LHCII remains associated with PSII (88), suggesting that the partitioning of LHCII between PSII and PSI is determined largely by changes in the binding affinity of LHCII to these complexes

through phosphorylation. Evidence for the migration of LHCII between PSII and PSI is based primarily on in vitro studies with thylakoid membranes (66, 120). Is there any direct in vivo evidence for this process? Using fluorescence lifetime imaging microscopy, which allows one to differentiate chlorophyll autofluorescence by its lifetime, Iwai et al. (60) were able to detect a 250-ps-lifetime fluorescence component in live *Chlamydomonas* cells during a transition from state 1 to state 2. This component was absent in an *stt7* mutant that is deficient in LHCII phosphorylation and unable to perform state transitions (36, 45); in this mutant, the component spread through the cell and formed large spotted areas. Interestingly, this component was also detected in a mutant that contained LHCII but lacked both PSI and PSII, and its appearance correlated with LHCII phosphorylation. These data suggest that the 250-ps component originates from phospho-LHCII dissociation from PSII in vivo. Moreover, phospho-LHCII aggregates formed in state 2 and were energy dissipative (60). Aggregates of LHCII also occur during nonphotochemical quenching (see below) (55), but how they relate to those formed during state transitions is unclear. These results suggest the existence of a free LHCII pool, but whether it forms only transiently and how and where LHCII binds to PSI are also unclear. Clearly, this novel fluorescence imaging technique appears to be very promising for studying the dynamics of LHCII in the thylakoid membranes.

ROLE OF THE STT7/STN7 KINASE AND PPH1/TAP38 PHOSPHATASE IN STATE TRANSITIONS

Shortly after the discovery of state transitions, Bennett (16) detected LHCII kinase activity associated with the thylakoid membrane. However, attempts to isolate this kinase using biochemical methods failed (30, 31, 117). *Chlamydomonas* proved to be particularly well suited for a genetic screen of LHCII kinase mutants because state transitions in this alga are accompanied by large fluorescence changes that can be easily monitored by fluorescence video imaging (45, 76). This approach led to the isolation of *stt7*, a mutant that is blocked in state 1 and deficient in LHCII phosphorylation under state 2 conditions and that lacks the Stt7 protein kinase (36). This kinase is related to another thylakoid kinase, dubbed Stt1. Moreover, both Stt7 and Stt1 have orthologs in *Arabidopsis*, called STN7 (involved in LHCII phosphorylation and state transitions) and STN8 (required for the phosphorylation of the PSII core proteins), respectively (14, 21, 123). These kinases are also present in other algae and land plants.

Although it is clear that the Stt7/STN7 kinase is required for the transition from state 1 to state 2 and for LHCII phosphorylation, it has not yet been proven that LHCII is its direct substrate. Nevertheless, this is likely to be the case, because among the other characterized chloroplast kinases, none is specifically required for LHCII phosphorylation. A comparative phosphoproteomic study of *Chlamydomonas* thylakoid membranes from wild-type and *stt7* plants under state 1 and state 2 conditions revealed several Stt7-dependent phosphorylation sites in the N-terminal stromal regions of Lhcbm1/Lhcbm10, Lhcbm4/Lhcbm6/Lhcbm8/Lhcbm9, Lhcbm3, Lhcbm5, and CP29 and on the Stt1 kinase, suggesting functional interactions between these two kinases (80). Stt7/STN7 protein could also be detected in phosphorylated form (80, 109). Although the functional significance of this phosphorylation is not known in *Chlamydomonas*, in *Arabidopsis* the decrease of the STN7 kinase level under state 1 conditions appears to be linked to the dephosphorylation of STN7 (128).

A transmembrane domain separates the N-terminal luminal region of Stt7 from its catalytic domain on the stromal side of the thylakoid membrane (81). The kinase appears to form a dimer (131). Two conserved cysteine residues near the N terminus are essential for the activity of the kinase, although they are separated from the catalytic kinase domain. They could be involved in the dimerization of Stt7. The kinase is firmly associated with *Cytb₆f* but in substoichiometric amounts,

with a ratio of 1:20 (81). Although the activation of Stt7 is known to depend on binding of PQH₂ to the Q_o site of *Cytb₆f*, it is not clear how the signal from the Q_o site on the lumen side is transmitted to the catalytic domain on the stromal side of the thylakoid membrane. Coimmunoprecipitation experiments revealed that, in addition to its association with *Cytb₆f*, Stt7 interacts with LHCII and PSI but not with PSII (81).

The Stt7/STN7 kinase is required not only for state transitions but also for proper acclimation to fluctuations in white-light intensity. *Arabidopsis* plants subjected to alternating short periods of low and high light undergo reversible LHCII protein phosphorylation (121). Under these conditions, in low light the STN7 kinase is activated and phosphorylates LHCII, and in high light the STN7 kinase is inactivated and LHCII is dephosphorylated, presumably by the PPH1/TAP38 phosphatase (108, 114). Surprisingly, these reversible changes in LHCII phosphorylation are not accompanied by changes in the relative excitation of the two photosystems, in contrast to what occurs during state transitions, when the two photosystems are excited artificially by light preferentially absorbed by PSII or PSI (121). A challenging task will be to elucidate how the STN7 kinase together with LHCII phosphorylation is able to maintain the redox poise of the electron transport chain under fluctuating light without any apparent adjustment of the antenna cross sections of both photosystems.

Among the dozens of *Chlamydomonas* state-transition mutants, none was affected in a protein phosphatase involved in LHCII dephosphorylation during a transition from state 2 to state 1. In contrast, a coupled bioinformatics-biochemical approach in *Arabidopsis* proved more successful. This approach involved screening homozygous mutant lines with T-DNA insertions in genes of predicted chloroplast phosphatases and a dephosphorylation test on young seedlings during a transition from state 2 to state 1 (108, 114). In this way, a protein phosphatase 2C dubbed PPH1/TAP38 was identified that is specifically required for the dephosphorylation of LHCII. The same reverse-genetic screen also identified the PSII core phosphatase (PBCP) for the dephosphorylation of the PSII core proteins (112). Thus, PPH1 and PBCP represent the counterparts of the STN7 and STN8 protein kinases, and together these proteins are the major determinants of the phosphorylation status of the LHCII and PSII core proteins. Although there is clear evidence for a redox control of STN7, whether the activity of the two identified phosphatases is regulated or whether these enzymes are constitutively active is unknown.

REGULATION OF LIGHT-HARVESTING COMPLEX GENE EXPRESSION AND ASSEMBLY

Photosynthetic organisms acclimate to changes in light quantity and quality through compensatory responses in which changes in LHCII gene expression lead to adjustments of antenna size. Because the LHC genes are in the nucleus, coupling of nuclear gene expression with light sensing in the chloroplast is required in order to elicit a proper response. Regulation of LHC gene expression occurs at the transcriptional (89), posttranscriptional (38, 44, 47), and translational level (46, 99). In many cases, the size of the light-harvesting systems of algae and plants is enlarged under low irradiance to improve light capture by PSII and PSI but is reduced under high irradiance to avoid overexcitation of the photosystems and thereby to prevent photooxidative damage.

Expression of the LHC genes is affected not only by light but also by other factors and processes, including temperature, circadian rhythm (90), reactive oxygen species, and tetrapyrrole intermediates (62). The underlying mechanisms of regulation are still poorly understood, although some of the components of the signaling chains through which the nucleus perceives metabolic changes in the chloroplast have been identified (for a review, see 104). A key factor in these signaling events is the redox state of the photosynthetic electron transport chain, in particular that of the PQ pool,

SRP: signal
recognition particle

which depends on the relative excitation of PSII and PSI. One potential candidate for sensing this redox state is the Stt7/STN7 kinase, which is involved not only in short-term responses such as state transitions but also in a long-term response that involves signaling to the nucleus. Analysis of several *Arabidopsis* mutants deficient in peripheral PSI subunits revealed that, although they are deficient in state transitions, the long-term response is not affected (107); this indicates that state transitions are not required for the long-term response and that the branch point between state transitions and the long-term response is downstream of STN7. However, the downstream components of these signaling chains are still largely unknown.

One surprising feature of the antenna systems of PSII and PSI is that they are regulated in different ways. As an example, upon acclimation of *Arabidopsis* plants from low to high light, the PSII antenna size is greatly diminished. Although the amounts of Lhcb4 (CP29) and Lhcb5 (CP26) do not change relative to the PSII reaction center, Lhcb1, Lhcb2, and Lhcb6 (CP24) are strongly downregulated (9). Because CP24 is a minor component and accounts for less than 5% of the total PSII chlorophyll, its decrease has only a modest effect on the PSII antenna size. This protein has a peripheral position in the PSII supercomplex and may play a role in stabilizing the binding of the LHCII trimers to PSII. In contrast, the stoichiometry of all Lhca antenna proteins with respect to the PSI core complex is unchanged under the same conditions (9). Recent studies suggest regulation at the translational level upon light stress (46). Whereas translation of Lhcb6 and Lhcb4.2 is downregulated, translation of Lhcb4.3 is upregulated, suggesting that the former subunits are involved in light harvesting and the latter has a photoprotective function. In PSI, in contrast, the amount of the Lhca proteins relative to the PSI reaction center remains constant upon exposure to high light; the light-harvesting function of PSI is instead regulated through changes in the ratio of PSI to PSII and the association of Lhcb proteins to PSI (9).

Several factors are involved in integrating and assembling the LHC proteins in the thylakoid membranes (Table 2), including the chloroplast signal recognition particle (SRP), consisting of cpSRP54 and cpSRP43, which interact posttranslationally with SRP's LHC substrates (71, 113). Together, these proteins form a transit complex required for keeping the hydrophobic LHC proteins soluble in the stroma during their migration from the envelope to the thylakoid membranes and for inserting the LHC proteins into these membranes. Assembly of the light-harvesting system requires Alb3, which is well conserved in land plants and algae. It is structurally related to the mitochondrial Oxa1p and bacterial YidC proteins, which are essential for integrating proteins into membranes (25, 87). Loss of this protein in *Arabidopsis* leads to an albino phenotype (118), and in vitro studies have shown that this protein is required for the insertion of the LHCII protein into thylakoid membranes (95). In *Chlamydomonas*, two nucleus-encoded proteins, Alb3.1 and Alb3.2, are related to Oxa1p. Loss of Alb3.1 leads to the specific depletion of LHCII and to a decrease of PSII (15). Alb3.2 appears to play an essential role in thylakoid membrane assembly, and a mild decrease of this protein is lethal (50).

A genetic screen for *Chlamydomonas* mutants with decreased antenna size revealed *Tla1*, a nuclear gene whose product acts as a regulator of chlorophyll content and chlorophyll antenna size and appears to be localized on the chloroplast envelope and/or budding envelope membrane (93, 94). This was further confirmed by overexpression and repression of *Tla1*, which led to a larger and smaller antenna, respectively. The thylakoid membranes were disorganized in the *tla1* mutant, with a loss of properly appressed grana membranes. The depletion of *Tla1* also affects accumulation of the PSII reaction center proteins D1, D2, and VIPP1, the last of which is a protein conserved in land plants, algae, and cyanobacteria that is involved in thylakoid membrane biogenesis or maintenance (93).

The cytoplasmic Nab1 protein in *Chlamydomonas* is another factor regulating antenna size. It acts as a cytoplasmic repressor of LHCII translation by binding selectively specific Lhcbm isoforms

Table 2 Regulatory components involved in light-harvesting complex (LHC) and/or thylakoid membrane assembly and signaling

| Protein | <i>Cr</i> | <i>At</i> | Activity/features | Biological function |
|--------------|-----------|-----------|----------------------------------|---|
| Stt7/STN7 | ✓ | ✓ | Kinase: LHCII | State transitions, retrograde signaling |
| Stl1/STN8 | ✓ | ✓ | Kinase: PSII core | PSII repair, thylakoid folding |
| Pph1/TAP38 | ✓ | ✓ | Phosphatase: LHCII | State transitions |
| Pbcp/PBCP | ✓ | ✓ | Phosphatase: PSII core | |
| cpSRP54 | ✓ | ✓ | | LHC insertion |
| cpSRP43 | ✓ | ✓ | | LHC insertion |
| Alb3.1 | ✓ | ✓ | | LHCII assembly |
| Alb3.2 | ✓ | | | Thylakoid assembly/biogenesis |
| Tla1 | ✓ | | | Thylakoid assembly |
| FZL | | ✓ | Dynammin-like GTPase | Thylakoid folding |
| Vipp1 | ✓ | ✓ | | Thylakoid biogenesis |
| Vipp2 | ✓ | | | Thylakoid biogenesis |
| CURT1a,b,c,d | ✓ | ✓ | | Thylakoid membrane curvature |
| GUN1 | | ✓ | Tetratricopeptide repeat protein | Retrograde signaling |

Abbreviations: *Cr*, *Cblamydomonas reinhardtii*; *At*, *Arabidopsis thaliana*; PSII, photosystem II. In the *Cr* and *At* columns, check marks indicate that a given protein is present in that species. In the activity/features and biological-function columns, blank cells indicate that those aspects are not clearly understood.

with Lhcbm6 mRNA as its main target, thereby sequestering the mRNA in translationally silent ribonucleoprotein complexes. Its activity is regulated both by cysteine-based redox control and by arginine methylation (18, 129). Apparently the increased or decreased need for LHCII protein synthesis is sensed by Nab1 through changes in the cytosolic redox state. However, little is known about how the plastid redox state influences the cytosolic redox state. Depletion of Tla1 does not affect the Nab1 level, suggesting that Tla1 and Nab1 act in different signaling pathways.

ENERGY-DEPENDENT NONPHOTOCHEMICAL QUENCHING

Upon light absorption by the antenna system, the excitation energy can be dissipated in several ways (73): It can be used for photochemistry, emitted as chlorophyll fluorescence, or converted into heat in the antenna complexes. Because this last process is associated with a decrease of fluorescence, it is referred to as nonphotochemical quenching of chlorophyll fluorescence (NPQ). Based on the kinetics of fluorescence relaxation in the dark, at least three different components of NPQ can be distinguished: the energy-dependent component, q_E , which depends on the proton gradient across the thylakoid membrane and relaxes within seconds; a second component, q_T , which is caused by state transitions and relaxes within minutes; and a third component, q_I , which is caused by photoinhibition and relaxes very slowly.

q_E -NPQ is triggered by the light-induced proton gradient across the thylakoid membrane. The acidification of the thylakoid lumen leads to the activation of violaxanthin de-epoxidase, which converts violaxanthin through de-epoxidation first to antheraxanthin and then to zeaxanthin, a series of reversible reactions that are part of the xanthophyll cycle (40, 132). The conversion of these xanthophylls, which are bound to the LHC polypeptides, induces conformational changes in these proteins. Remarkably, the LHC proteins are able to switch from an efficient light-harvesting

state to a photoprotective state in which the light excitation energy is thermally dissipated through subtle perturbations in their physicochemical environment (75). To identify which of the LHCII complexes are involved in qE-NPQ, *Arabidopsis* mutant plants were examined in which specific LHCII components were removed through antisense technique or knockouts. This analysis revealed that, whereas the absence of CP26 did not affect qE, the absence of CP29 and CP24 decreased qE by 30% and 50%, respectively (4, 72). Loss of the major Lhcb1 and Lhcb2 proteins decreased qE-NPQ by 35% (5). These results make it unlikely that an individual LHCII complex acts as the only site of qE; rather, they indicate that this process occurs at multiple sites within the LHCII antenna. Several mechanisms have been proposed, including excitonic coupling (19) and charge transfer (54) or energy transfer (110) between xanthophyll and chlorophyll molecules or chlorophyll–chlorophyll charge transfer states (91).

Genetic screens of mutants deficient in NPQ based on their altered fluorescence properties confirmed the importance of the xanthophyll cycle for NPQ (101, 102). Moreover, this approach also identified NPQ mutants unaffected in the xanthophyll cycle, leading to the discovery of PsbS, which belongs to the superfamily of LHC proteins but contains four rather than three transmembrane domains (84) and does not bind pigments (24). It acts as a sensor of the lumen pH and activates qE rapidly, presumably upon protonation of its acidic domains exposed to the thylakoid lumen, which are thought to promote a rearrangement of the PSII supercomplex in the grana region (17, 51). Although PsbS is clearly essential for the rapid induction of qE in vivo, it is dispensable in vitro, as qE can be induced without PsbS provided that the pH gradient is sufficiently high (63). This suggests that direct protonation of LHCII or of other thylakoid proteins can bypass the need for PsbS. Whether this property has any physiological significance remains to be seen.

Comparison of the light-harvesting systems of cyanobacteria, algae, and plants has revealed that these systems are the most variable part of the photosynthetic apparatus. It is therefore not surprising that different regulatory mechanisms have arisen during evolution, in particular with regard to qE. The case of *Chlamydomonas* is particularly interesting. As with land plants, this alga displays Δ pH- and xanthophyll-dependent qE. Although it contains a PsbS-like protein, another protein, LHCSR, is required for qE in this organism (106). This protein belongs to the LHC superfamily and is conserved in most photosynthetic eukaryote taxa except for red algae and vascular plants. Quenching of excitation energy occurs by a chlorophyll–carotenoid charge transfer mechanism in this protein (23). In contrast to PsbS, this protein binds several chlorophylls and xanthophylls. However, it has a very short chlorophyll fluorescence lifetime in vitro that is further shortened at low pH (23), suggesting that some quenching activity occurs even in low light, activity that is presumably increased by protonation of its lumen acidic domains under high light. This constitutive quenching under low light may be the reason that LHCSR gene expression is low under these conditions (to avoid wasteful energy dissipation) and upregulated in high light. LHCSR3 binds PSII presumably by interacting with other LHCII proteins, in particular Lhcbm1 present in the trimers of the PSII antenna (1). Based on mutant studies, Lhcbm1 is specifically required for qE in *Chlamydomonas* (39), indicating that although the major LHCII proteins have very similar sequences, they can have distinct functions. As with other LHCII proteins, LHCSR3 is phosphorylated by the Stt7/STN7 kinase under state 2 conditions and associates with PSI (23). It will be interesting to determine whether it can quench excitation energy in the free detached LHCII proteins and induce qE in PSI.

Allouret et al. (1) studied acclimation to high light in *Chlamydomonas* by comparing *npq4* and *stt7* single mutants and *npq4 stt7* double mutants. It takes several hours to fully induce LHCSR3 and hence qE-NPQ in *Chlamydomonas* upon exposure to high light; a transition from state 1 to state 2, in contrast, is induced within minutes under these conditions. The comparison of fluorescence quenching, photosynthetic activity, and reactive oxygen species formation in the three mutants

and in the wild type revealed that both qE-NPQ and state transitions are induced under high light. State transitions play an important role in this response, as they promote the transfer of the mobile LHCII from PSII to PSI, thus decreasing the excitation pressure on PSII as well as photodamage. qE-NPQ and state transitions therefore appear to act together in a complementary way during acclimation to high light: qE-NPQ constitutes the major response for photoprotection in a steady state, whereas state transitions prevent photodamage during the early response and may also contribute to photoprotection in a steady state. In this respect, it is interesting that in some microalgae, such as the diatom *Phaeodactylum tricornutum*, the LHCSR proteins responsible for qE-NPQ are constitutively expressed, but these organisms are unable to perform state transitions (105).

PHOTOSYSTEM II PROTEIN PHOSPHORYLATION AND THYLAKOID MEMBRANE FOLDING

The PSII core proteins D1, D2, and CP43 are phosphorylated by the Stl1/STN8 kinase, which forms a two-member family with the Stt7/STN7 kinase (36). The corresponding phosphatase of Stl1/STN8 is PBCP, which is specifically involved in dephosphorylating the PSII core proteins (112).

Thylakoid membrane organization in grana and stromal regions is shaped to a large extent by the resident photosynthetic complexes. Thus, mutants deficient in PSI contain mostly grana with a reduced amount of stroma lamella (11, 82). The protein density is high, especially in the grana regions, with 70% of the surface area occupied by proteins and 30% by lipids. This molecular crowding greatly limits the mobility of proteins within the grana, especially that of PSII and its light-harvesting system. Recent work indicates that light-induced architectural changes in the folding of thylakoid membranes occur that appear to be mediated in part by changes in phosphorylation of thylakoid proteins through the action of the thylakoid protein kinases Stt7/STN7 and Stl1/STN8 and the phosphatases PPH1/TAP38 and PBCP (48, 112). These changes may alleviate the limited mobility of proteins in these membranes.

At first sight, molecular crowding may be a problem for the PSII repair cycle, which involves protein migration within the grana region and operates as follows (**Figure 3**). PSII is prone to photodamage because it catalyzes the water-splitting reaction of photosynthesis, which is one of the strongest oxidizing reactions in living organisms. To maintain the photosynthetic yield of PSII, a highly efficient repair system has evolved (7, 67). Upon light-induced photodamage of PSII in the grana membrane, the complex moves from the grana to the stromal regions, where damaged D1, the most vulnerable PSII subunit, is degraded on both the stromal and luminal sides of the thylakoid membrane by the FtsH and Deg proteases. A newly synthesized D1 subunit is then inserted cotranslationally into the complex, and upon repair, PSII moves back to the grana region. This repair cycle is driven by successive phosphorylation and dephosphorylation of the PSII core proteins.

Interestingly, D1 degradation is significantly retarded in *stn8* single mutants and *stn7 stn8* double mutants of *Arabidopsis* (48), in which folding of the thylakoid membrane is affected. Loss of the STN8 kinase leads to increased grana size and fewer grana layers. This feature is particularly noteworthy because grana size is remarkably conserved in land plants and algae (68). Moreover, in the absence of STN8, the partitioning of FtsH between the grana and stromal membranes changes (48). The access of FtsH to the grana is restricted and the movement of damaged D1 from the grana to the stromal membranes is hindered, suggesting that the phosphorylation of the PSII core proteins is important for facilitating protein mobility within the thylakoid membranes. This view is further supported by studies of the protein phosphatase PBCP, which is involved in dephosphorylating the PSII core proteins and represents the counterpart of the STN8 kinase (112).

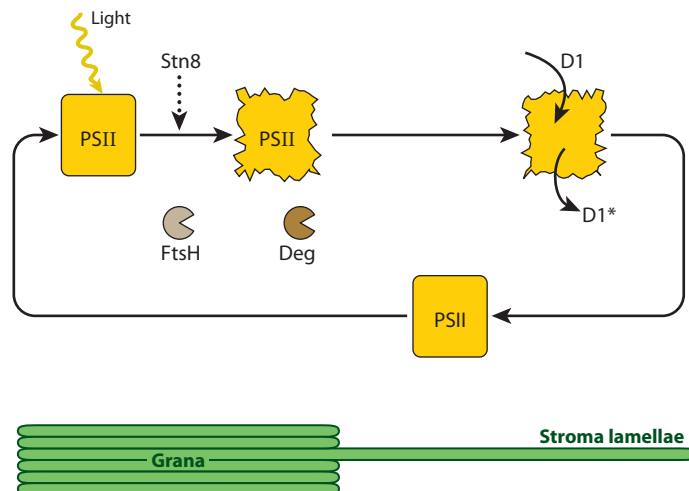


Figure 3

Photosystem II (PSII) repair cycle. Upon illumination with high light, the PSII reaction center—especially the D1 protein—is progressively damaged. The PSII core proteins are phosphorylated by the Stt1/STN8 kinase, and the damaged complex migrates from the grana to the stroma lamellae. The D1 protein is degraded by the FtsH and Deg proteases, and upon its removal from the PSII reaction center, a newly synthesized D1 protein is inserted cotranslationally into the complex, which moves back to the grana and thereby completes the repair cycle. D1* denotes released D1 fragments.

Arabidopsis mutants lacking this phosphatase have fewer layers in the grana stacks compared with the wild type.

It is interesting that exposure of thylakoids to high light induces two major structural changes that act synergistically to facilitate the migration of PSII in the grana to its repair machinery in the stroma lamellae (52). First, lateral shrinkage of the granum diameter from approximately 370 to 300 nm occurs, which reduces the diffusion distance of damaged PSII in the grana to the stroma lamellae and thus provides PSII with easier access to FtsH. This change also increases the ratio of granum perimeter to area, thereby increasing the contact zones between appressed and unappressed membranes. Second, protein mobility is increased in the high-light-exposed thylakoid membranes. These conditions also induce phosphorylation of the PSII core proteins. Taken together, these results strongly suggest that phosphorylation of the PSII core proteins significantly impacts the folding and architecture of the thylakoid membranes. As suggested in earlier studies, protein phosphorylation of major thylakoid proteins could induce electrostatic repulsion between the grana membrane layers and thereby loosen the grana structure (10).

Analysis of *Arabidopsis* mutants in which the thylakoid membrane system is perturbed has identified additional proteins required for proper folding of thylakoid membranes. One of these studies revealed FZL, an FZO-like chloroplast protein, as a determinant of chloroplast morphology and thylakoid ultrastructure (49). This protein belongs to the dynamin superfamily of membrane-remodeling GTPases and is associated with the thylakoid and envelope membranes. Mutants lacking FZL display disorganized grana stacks and changes in the relative proportion of grana and stromal thylakoids. As FZO proteins are known to promote mitochondrial fusion in animals, it is possible that the chloroplast protein participates in membrane remodeling, perhaps through vesicle trafficking from the inner envelope for thylakoid formation. It is surprising, however, that in spite of the perturbed thylakoid membrane architecture in the *fzl* mutant, state transitions are not affected (M. Tikkanen & J.D. Rochaix, unpublished observations).

The chloroplast protein VIPP1 has also been proposed to function in thylakoid membrane biogenesis and assembly (74). VIPP1 was originally identified as a potential lipid transfer protein in the chloroplast envelope (83) and is conserved in vascular plants, algae, and cyanobacteria. Further studies indicated that it is localized in both the envelope and thylakoids and is involved in thylakoid biogenesis through vesicle formation (8, 74). Accumulation of vesicles thought to give rise to thylakoid membranes has also been observed in the nongreen sectors of a variegated *thf1* mutant of *Arabidopsis* that lacks the plastid THF1 protein and has a disorganized thylakoid membrane system (125). In *Chlamydomonas*, Vipp1 interacts with the chloroplast chaperones Cdj2, Hsp70, and Hsp90, which catalyze the assembly and disassembly of Vipp1 oligomers (85). The role of these oligomers in thylakoid membrane assembly is not clear, however. *Chlamydomonas* has two paralogs, Vipp1 and Vipp2, which appear to be redundantly involved in the biogenesis/assembly of thylakoid membrane core complexes, perhaps by delivering small hydrophobic proteins or lipids (103). However, in neither case is it clear how or whether these different proteins act in the same pathway.

One conspicuous feature of appressed grana membranes is the extreme curvature of the lipid bilayer at their margins. Recent work has identified a chloroplast curvature thylakoid 1 (Curt1) protein family of four members involved in this process (6). These proteins are conserved in plants, algae, and cyanobacteria and modulate thylakoid architecture by inducing membrane curvature. In vitro Curt1 proteins induce tubulation of liposomes. These proteins are localized mainly at grana margins and form large oligomers. Grana architecture could be correlated with the amount of the Curt1 proteins (6). Overexpression of these proteins leads to the formation of an increased number of grana membrane layers with a smaller granum diameter. In contrast, mutants lacking these proteins form larger grana discs with only a few grana layers and margins. Interestingly, the thylakoid membrane architecture observed when Curt1a accumulates to 50% relative to wild-type levels resembles that of the *stn7 stn8* double mutant, in which PSII core and LHCII phosphorylation is strongly reduced. However, the fact that phosphorylation of these proteins in the *curt1* mutants is induced and occurs in both the dark and the light suggests that the loss of the Curt proteins overrides the effect of phosphorylation and that PSII core protein phosphorylation does not play any significant role in thylakoid curvature mediated by Curt1 proteins (6). Interestingly, phosphoproteomic studies indicate that some Curt proteins are among the substrates of the STN8 kinase (48). Whether the activity of the Curt1 proteins is regulated through phosphorylation and to what extent this could thereby control thylakoid membrane folding remain to be seen.

PERSPECTIVES

The picture that emerges from recent studies on the thylakoid membrane is of a highly dynamic system in which structural rearrangements are triggered by changes in light conditions. The light-harvesting system of PSII plays a key role in these processes, as it acts both as a light-energy collector to drive photosynthesis and as a sensor and energy dissipator when the absorbed energy exceeds the capacity of the photosynthetic apparatus. All photosynthetic organisms face the challenge of regulating light harvesting under fluctuating environmental conditions. Complex signaling pathways within the chloroplast and from the chloroplast to the nucleus have evolved that perceive changes in the light environment and trigger cellular responses to restore the redox poise of the photosynthetic electron transport chain. Most prominent among the identified components of these signaling chains is a quartet consisting of two protein kinases (Stt7/STN7 and Stt1/STN8) and their respective counterpart phosphatases (PPH1/TAP38 and PBCP), which determine the phosphorylation status of the PSII light-harvesting system and the PSII core complex. Although

these enzymes have been examined in some detail, their mode of activation and their downstream targets are still largely unknown and represent important areas of future research.

New insights have arisen from a comparative analysis of acclimation and photoprotection in algae and land plants. These studies have revealed different primary ΔpH sensors: LHCSR in algae functions both as a sensor of excess light and as a site of quenching, whereas PSBS in plants acts only as a sensor, with multiple sites of quenching in the LHC antenna proteins. Single-molecule studies have demonstrated that LHC proteins can dynamically switch from a light-harvesting to a quenching state (75), and understanding the structural basis of this switch remains a challenging task. Cryptophytes and peridinin-containing dinoflagellates accomplish qE, although they lack both LHCSR and PSBS; it will be interesting to elucidate how quenching occurs in these organisms. Two other differences between algae and plants are the size of the mobile LHCII antenna and the extent of state transitions, which are considerably larger in algae than in land plants. These organisms diverged several hundred million years ago and adapted to their specific habitats. Although the exact reasons for these differences are not clear, one possibility is that because induction of qE-NPQ is slow in algae, additional photoprotection during this critical period may be provided by state transitions (1). More generally, a larger capacity for state transitions could confer a higher degree of flexibility to adapt to stress conditions other than high light, such as CO_2 limitation and nutrient deprivation, and to metabolic changes that maintain an adequate ATP/NADPH ratio (130).

Modeling of photosynthesis has shown that an increase in the rate of recovery from NPQ under fluctuating light conditions could significantly improve the photosynthetic yield when the light is limiting (98, 133). Thus, further mechanistic studies of qE are needed in order to design new strategies aimed at optimizing photoprotection and plant growth.

We are still at an early stage of understanding how thylakoid membranes are formed and folded, although a few factors involved in these processes have been identified. The structural arrangement of this membrane network in grana and stroma lamellae is highly dynamic in response to changes in the light environment. It involves complex phosphorylation and dephosphorylation events of major thylakoid proteins and concerted migration of proteins and protein complexes in a highly crowded membrane environment, a process that is still poorly understood and that will require new approaches based on protein tagging and innovative fluorescence microscopy techniques such as fluorescence correlation microscopy, which allows one to determine the diffusion of single molecules in the thylakoid membrane (60). There is little doubt that research on this system will continue to be challenging and exciting.

SUMMARY POINTS

1. Photosynthetic organisms are subjected to continuous changes in light intensity and spectral quality and must adjust their photosynthetic apparatus for optimal performance and photoprotection.
2. Absorbed light energy can be used for photosynthesis, emitted as fluorescence, or dissipated as heat when the capacity of the photosynthetic apparatus is saturated.
3. The light-harvesting system of photosystem II acts both as a light-energy collector under limiting light and as an energy dissipator under excess light.
4. Thermal dissipation of excess excitation energy occurs through energy-dependent non-photochemical quenching, whereby acidification of the thylakoid lumen induces conformational changes in the light-harvesting proteins.

5. Balancing of the light energy between the two photosystems occurs through state transitions, a process in which the mobile part of the photosystem II antenna moves between photosystem II in the grana and photosystem I in the stroma lamellae to restore the redox poise whenever the relative excitation of the two photosystems changes.
6. The mobility of the light-harvesting system of photosystem II is regulated by phosphorylation, which in turn is controlled by the redox state of the plastoquinone pool.
7. A quartet consisting of two protein kinases (Stt7/STN7 and Stt1/STN8) and their respective counterpart phosphatases (PPH1/TAP38 and PBCP) determines the phosphorylation status of the light-harvesting complex II and photosystem II core proteins.
8. Because of its intrinsic photochemical activity, photosystem II is prone to photodamage and is continuously repaired through a process that involves reversible phosphorylation of the core subunits of photosystem II, migration of the damaged complex from the grana to the stroma lamellae for repair, and the complex's return to the grana.

DISCLOSURE STATEMENT

The author is not aware of any affiliations, memberships, funding, or financial holdings that might be perceived as affecting the objectivity of this review.

ACKNOWLEDGMENTS

I thank Nicolas Roggli for preparing the figures and Michel Goldschmidt-Clermont and Geoffrey Fucile for critical comments. Work in my laboratory was supported by grant 31003A_133089/1 from the Swiss National Foundation.

LITERATURE CITED

1. Alloreant G, Tokutsu R, Roach T, Peers G, Cardol P, et al. 2013. A dual strategy to cope with high light in *Chlamydomonas reinhardtii*. *Plant Cell* 25:545–57
2. Amunts A, Drory O, Nelson N. 2007. The structure of a plant photosystem I supercomplex at 3.4 Å resolution. *Nature* 447:58–63
3. Andersson B, Andersson J. 1980. Lateral heterogeneity in the distribution of chlorophyll-protein complexes of the thylakoid membranes of spinach chloroplasts. *Biochem. Biophys. Acta* 593:427–40
4. Andersson J, Walters RG, Horton P, Jansson S. 2001. Antisense inhibition of the photosynthetic antenna proteins CP29 and CP26: implications for the mechanism of protective energy dissipation. *Plant Cell* 13:1193–204
5. Andersson J, Wentworth M, Walters RG, Howard CA, Ruban AV, et al. 2003. Absence of the Lhcb1 and Lhcb2 proteins of the light-harvesting complex of photosystem II—effects on photosynthesis, grana stacking and fitness. *Plant J.* 35:350–61
6. Armbruster U, Labs M, Pribil M, Viola S, Xu W, et al. 2013. *Arabidopsis* CURVATURE THYLAKOID1 proteins modify thylakoid architecture by inducing membrane curvature. *Plant Cell* 25:2661–78
7. Aro EM, Suorsa M, Rokka A, Allahverdiyeva Y, Paakkarinen V, et al. 2005. Dynamics of photosystem II: a proteomic approach to thylakoid protein complexes. *J. Exp. Bot.* 56:347–56
8. Aseeva E, Ossenbuhl F, Eichacker LA, Wanner G, Soll J, Voithknecht UC. 2004. Complex formation of Vipp1 depends on its α -helical PspA-like domain. *J. Biol. Chem.* 279:35535–41
9. Ballottari M, Dall'Osto L, Morosinotto T, Bassi R. 2007. Contrasting behavior of higher plant photosystem I and II antenna systems during acclimation. *J. Biol. Chem.* 282:8947–58

10. Barber J. 1982. Influence of surface charges on thylakoid structure and function. *Annu. Rev. Plant Physiol.* 33:261–95
11. Barneche F, Winter V, Crevecoeur M, Rochaix JD. 2006. ATAB2 is a novel factor in the signalling pathway of light-controlled synthesis of photosystem proteins. *EMBO J.* 25:5907–18
12. Bassi R, Giacometti GM, Simpson DJ. 1988. Changes in the organization of stroma membranes induced by in vivo state 1–state 2 transition. *Biochim. Biophys. Acta* 935:152–65
13. Bassi R, Machold O, Simpson DJ. 1985. Chlorophyll-proteins of two photosystem I preparations from maize. *Carlsberg Res. Commun.* 50:145–62
14. Bellafiore S, Barneche F, Peltier G, Rochaix JD. 2005. State transitions and light adaptation require chloroplast thylakoid protein kinase STN7. *Nature* 433:892–95
15. Bellafiore S, Ferris P, Naver H, Gohre V, Rochaix JD. 2002. Loss of Albino3 leads to the specific depletion of the light-harvesting system. *Plant Cell* 14:2303–14
16. Bennett J. 1979. Chloroplast phosphoproteins: phosphorylation of polypeptides of the light-harvesting chlorophyll protein complex. *Eur. J. Biochem.* 99:133–37
17. Betterle N, Ballottari M, Zorzan S, de Bianchi S, Cazzaniga S, et al. 2009. Light-induced dissociation of an antenna hetero-oligomer is needed for non-photochemical quenching induction. *J. Biol. Chem.* 284:15255–66
18. Blifernéz O, Wobbe L, Niehaus K, Kruse O. 2011. Protein arginine methylation modulates light-harvesting antenna translation in *Chlamydomonas reinhardtii*. *Plant J.* 65:119–30
19. Bode S, Quentmeier CC, Liao PN, Hafi N, Barros T, et al. 2009. On the regulation of photosynthesis by excitonic interactions between carotenoids and chlorophylls. *Proc. Natl. Acad. Sci. USA* 106:12311–16
20. Boekema EJ, Hankamer B, Bald D, Kruip J, Nield J, et al. 1995. Supramolecular structure of the photosystem II complex from green plants and cyanobacteria. *Proc. Natl. Acad. Sci. USA* 92:175–79
21. Bonardi V, Pesaresi P, Becker T, Schleiff E, Wagner R, et al. 2005. Photosystem II core phosphorylation and photosynthetic acclimation require two different protein kinases. *Nature* 437:1179–82
22. Bonaventura C, Myers J. 1969. Fluorescence and oxygen evolution from *Chlorella pyrenoidosa*. *Biochim. Biophys. Acta* 189:366–83
23. Bonente G, Ballottari M, Truong TB, Morosinotto T, Ahn TK, et al. 2011. Analysis of LhcSR3, a protein essential for feedback de-excitation in the green alga *Chlamydomonas reinhardtii*. *PLoS Biol.* 9:e1000577
24. Bonente G, Howes BD, Caffarri S, Smulevich G, Bassi R. 2008. Interactions between the photosystem II subunit PsbS and xanthophylls studied in vivo and in vitro. *J. Biol. Chem.* 283:8434–45
25. Bonnefoy N, Chalvet F, Hamel P, Slonimski PP, Dujardin G. 1994. *OXA1*, a *Saccharomyces cerevisiae* nuclear gene whose sequence is conserved from prokaryotes to eukaryotes controls cytochrome oxidase biogenesis. *J. Mol. Biol.* 239:201–12
26. Bulté L, Gans P, Rebeille F, Wollman FA. 1990. ATP control on state transitions in *Chlamydomonas*. *Biochim. Biophys. Acta* 1020:72–80
27. Caffarri S, Kouril R, Kereiche S, Boekema EJ, Croce R. 2009. Functional architecture of higher plant photosystem II supercomplexes. *EMBO J.* 28:3052–63
28. Chow WS, Miller C, Anderson JM. 1991. Surface charges, the heterogeneous lateral distribution of the two photosystems, and thylakoid stacking. *Biochim. Biophys. Acta* 1057:69–77
29. Cleland RE, Bendall DS. 1992. Photosystem I cyclic electron transport: measurement of ferredoxin-plastoquinone reductase activity. *Photosynth. Res.* 34:409–18
30. Coughlan SJ, Hind G. 1986. Purification and characterization of a membrane-bound protein kinase from spinach thylakoids. *J. Biol. Chem.* 261:11378–85
31. Coughlan SJ, Hind G. 1987. Phosphorylation of thylakoid proteins by a purified kinase. *J. Biol. Chem.* 262:8402–8
32. Croce R, Canino G, Ros F, Bassi R. 2002. Chromophore organization in the higher-plant photosystem II antenna protein CP26. *Biochemistry* 41:7334–43
33. DalCorso G, Pesaresi P, Masiero S, Aseeva E, Schunemann D, et al. 2008. A complex containing PGRL1 and PGR5 is involved in the switch between linear and cyclic electron flow in *Arabidopsis*. *Cell* 132:273–85
34. Daum B, Nicastro D, Austin J II, McIntosh JR, Kuhlbrandt W. 2010. Arrangement of photosystem II and ATP synthase in chloroplast membranes of spinach and pea. *Plant Cell* 22:1299–312

35. Delosme R, Olive J, Wollman FA. 1996. Changes in light energy distribution upon state transitions: an in vivo photoacoustic study of the wild type and photosynthesis mutants from *Cblamydomonas reinhardtii*. *Biochim. Biophys. Acta* 1273:150–58
36. Depège N, Bellaïf S, Rochaix JD. 2003. Role of chloroplast protein kinase Stt7 in LHCII phosphorylation and state transition in *Cblamydomonas*. *Science* 299:1572–75
37. Drop B, Webber-Birungi M, Yadav SK, Filipowicz-Szymanska A, Fusetti F, et al. 2013. Light-harvesting complex II (LHCII) and its supramolecular organization in *Cblamydomonas reinhardtii*. *Biochim. Biophys. Acta* 1837:63–72
38. Durnford DG, Price JA, McKim SM, Sarchfield ML. 2003. Light-harvesting complex gene expression is controlled by both transcriptional and post-transcriptional mechanisms during photoacclimation in *Cblamydomonas reinhardtii*. *Physiol. Plant.* 118:193–205
39. Elrad D, Niyogi KK, Grossman AR. 2002. A major light-harvesting polypeptide of photosystem II functions in thermal dissipation. *Plant Cell* 14:1801–16
40. Eskling M, Avidsson PO, Akerlund HE. 1997. The xanthophyll cycle, its regulation and components. *Physiol. Plant.* 100:806–16
41. Fernyhough P, Foyer C, Horton P. 1983. The influence of metabolic state on the level of phosphorylation of the light-harvesting chlorophyll-protein complex in chloroplasts isolated from maize mesophyll. *Biochim. Biophys. Acta* 725:155–61
42. Finazzi G, Furia A, Barbagallo RP, Forti G. 1999. State transitions, cyclic and linear electron transport and photophosphorylation in *Cblamydomonas reinhardtii*. *Biochim. Biophys. Acta* 1413:117–29
43. Finazzi G, Rappaport F, Furia A, Fleischmann M, Rochaix JD, et al. 2002. Involvement of state transitions in the switch between linear and cyclic electron flow in *Cblamydomonas reinhardtii*. *EMBO Rep.* 3:280–85
44. Flachmann R, Kuhlbrandt W. 1995. Accumulation of plant antenna complexes is regulated by post-transcriptional mechanisms in tobacco. *Plant Cell* 7:149–60
45. Fleischmann MM, Ravel S, Delosme R, Olive J, Zito F, et al. 1999. Isolation and characterization of photoautotrophic mutants of *Cblamydomonas reinhardtii* deficient in state transition. *J. Biol. Chem.* 274:30987–94
46. Floris M, Bassi R, Robaglia C, Alboresi A, Lanet E. 2013. Post-transcriptional control of light-harvesting genes expression under light stress. *Plant Mol. Biol.* 82:147–54
47. Frigerio S, Campoli C, Zorzan S, Fantoni LI, Crosatti C, et al. 2007. Photosynthetic antenna size in higher plants is controlled by the plastoquinone redox state at the post-transcriptional rather than transcriptional level. *J. Biol. Chem.* 282:29457–69
48. Fristedt R, Willig A, Granath P, Crèvecoeur M, Rochaix JD, Vener A. 2009. Phosphorylation of photosystem II controls functional macroscopic folding of plant photosynthetic membranes in *Arabidopsis*. *Plant Cell* 21:3950–64
49. Gao H, Sage TL, Osteryoung KW. 2006. FZL, an FZO-like protein in plants, is a determinant of thylakoid and chloroplast morphology. *Proc. Natl. Acad. Sci. USA* 103:6759–64
50. Gohre V, Ossenhuhl F, Crèvecoeur M, Eichacker LA, Rochaix JD. 2006. One of two alb3 proteins is essential for the assembly of the photosystems and for cell survival in *Cblamydomonas*. *Plant Cell* 18:1454–66
51. Goral TK, Johnson MP, Duffy CD, Brain AP, Ruban AV, Mullineaux CW. 2012. Light-harvesting antenna composition controls the macrostructure and dynamics of thylakoid membranes in *Arabidopsis*. *Plant J.* 69:289–301
52. Herbstova M, Tietz S, Kinzel C, Turkina MV, Kirchhoff H. 2012. Architectural switch in plant photosynthetic membranes induced by light stress. *Proc. Natl. Acad. Sci. USA* 109:20130–35
53. Hertle AP, Blunder T, Wunder T, Pesaresi P, Pribil M, et al. 2013. PGRL1 is the elusive ferredoxin-plastoquinone reductase in photosynthetic cyclic electron flow. *Mol. Cell* 49:511–23
54. Holt NE, Zigmantas D, Valkunas L, Li XP, Niyogi KK, Fleming GR. 2005. Carotenoid cation formation and the regulation of photosynthetic light harvesting. *Science* 307:433–36
55. Horton P. 2012. Optimization of light harvesting and photoprotection: molecular mechanisms and physiological consequences. *Philos. Trans. R. Soc. B* 367:3455–65
56. Horton P, Black MT. 1980. Activation of adenosine 5' triphosphate-induced quenching of chlorophyll fluorescence by reduced plastoquinone. *FEBS Lett.* 119:141–45

57. Horton P, Ruban A. 2005. Molecular design of the photosystem II light-harvesting antenna: photosynthesis and photoprotection. *J. Exp. Bot.* 56:365–73
58. Iwai M, Takahashi Y, Minagawa J. 2008. Molecular remodeling of photosystem II during state transitions in *Chlamydomonas reinhardtii*. *Plant Cell* 20:2177–89
59. Iwai M, Takizawa K, Tokutsu R, Okamuro A, Takahashi Y, Minagawa J. 2010. Isolation of the elusive supercomplex that drives cyclic electron flow in photosynthesis. *Nature* 464:1210–13
60. Iwai M, Yokono M, Inada N, Minagawa J. 2010. Live-cell imaging of photosystem II antenna dissociation during state transitions. *Proc. Natl. Acad. Sci. USA* 107:2337–42
61. Jansson S. 1998. A guide to the LHC genes and their relatives in *Arabidopsis*. *Trends Plant Sci.* 4:236–40
62. Johanningmeier U, Howell SH. 1984. Regulation of light-harvesting chlorophyll-binding protein mRNA accumulation in *Chlamydomonas reinhardtii*: possible involvement of chlorophyll synthesis precursors. *J. Biol. Chem.* 259:13541–49
63. Johnson MP, Ruban AV. 2011. Restoration of rapidly reversible photoprotective energy dissipation in the absence of PsbS protein by enhanced Δ pH. *J. Biol. Chem.* 286:19973–81
64. Joliot P, Joliot A. 2006. Cyclic electron flow in C3 plants. *Biochim. Biophys. Acta* 1757:362–68
65. Kargul J, Nield J, Barber J. 2003. Three-dimensional reconstruction of a light-harvesting complex I-photosystem I (LHCI-PSI) supercomplex from the green alga *Chlamydomonas reinhardtii*: insights into light harvesting for PSI. *J. Biol. Chem.* 278:16135–41
66. Kargul J, Turkina MV, Nield J, Benson S, Vener AV, Barber J. 2005. Light-harvesting complex II protein CP29 binds to photosystem I of *Chlamydomonas reinhardtii* under State 2 conditions. *FEBS J.* 272:4797–806
67. Kato Y, Sakamoto W. 2009. Protein quality control in chloroplasts: a current model of D1 protein degradation in the photosystem II repair cycle. *J. Biochem.* 146:463–69
68. Kirchhoff H. 2008. Molecular crowding and order in photosynthetic membranes. *Trends Plant Sci.* 13:201–7
69. Kirchhoff H. 2013. Architectural switches in plant thylakoid membranes. *Photosynth. Res.* 116:481–87
70. Kirchhoff H, Hall C, Wood M, Herbstova M, Tsabari O, et al. 2011. Dynamic control of protein diffusion within the granal thylakoid lumen. *Proc. Natl. Acad. Sci. USA* 108:20248–53
71. Klimyuk VI, Persello-Cartieaux F, Havaux M, Contard-David P, Schuenemann D, et al. 1999. A chromo-domain protein encoded by the *Arabidopsis* *CAO* gene is a plant-specific component of the chloroplast signal recognition particle pathway that is involved in LHCP targeting. *Plant Cell* 11:87–99
72. Kovacs L, Damkjaer J, Kereiche S, Iliaia C, Ruban AV, et al. 2006. Lack of the light-harvesting complex CP24 affects the structure and function of the grana membranes of higher plant chloroplasts. *Plant Cell* 18:3106–20
73. Krause GH, Weiss E. 1991. Chlorophyll fluorescence and photosynthesis: the basics. *Annu. Rev. Plant Physiol. Plant Mol. Biol.* 42:313–49
74. Kroll D, Meierhoff K, Bechtold N, Kinoshita M, Westphal S, et al. 2001. *VIPP1*, a nuclear gene of *Arabidopsis thaliana* essential for thylakoid membrane formation. *Proc. Natl. Acad. Sci. USA* 98:4238–42
75. Kruger TP, Iliaia C, Johnson MP, Ruban AV, Papagiannakis E, et al. 2012. Controlled disorder in plant light-harvesting complex II explains its photoprotective role. *Biophys. J.* 102:2669–76
76. Kruse O, Nixon PJ, Schmid GH, Mullineaux CW. 1999. Isolation of state transition mutants of *Chlamydomonas reinhardtii* by fluorescence video imaging. *Photosynth. Res.* 61:43–51
77. Kyle DJ, Staehelin LA, Arntzen CJ. 1983. Lateral mobility of the light-harvesting complex in chloroplast membranes controls excitation energy distribution in higher plants. *Arch. Biochem. Biophys.* 222:527–41
78. Lam E, Ortiz W, Malkin R. 1984. Chlorophyll *a/b* proteins of photosystem I. *FEBS Lett.* 168:10–14
79. Lemeille S, Rochaix JD. 2010. State transitions at the crossroad of thylakoid signalling pathways. *Photosynth. Res.* 106:33–46
80. Lemeille S, Turkina MV, Vener AV, Rochaix JD. 2010. Stt7-dependent phosphorylation during state transitions in the green alga *Chlamydomonas reinhardtii*. *Mol. Cell. Proteomics* 9:1281–95
81. Lemeille S, Willig A, Depège-Fargeix N, Delessert C, Bassi R, Rochaix JD. 2009. Analysis of the chloroplast protein kinase Stt7 during state transitions. *PLoS Biol.* 7:e45
82. Lezhneva L, Meurer J. 2004. The nuclear factor HCF145 affects chloroplast *psaA-psaB-rps14* transcript abundance in *Arabidopsis thaliana*. *Plant J.* 38:740–53

83. Li HM, Kaneko Y, Keegstra K. 1994. Molecular cloning of a chloroplastic protein associated with both the envelope and thylakoid membranes. *Plant Mol. Biol.* 25:619–32
84. Li XP, Björkman O, Shih C, Grossman AR, Rosenquist M, et al. 2000. A pigment-binding protein essential for regulation of photosynthetic light harvesting. *Nature* 403:391–95
85. Liu C, Willmund F, Whitelegge JP, Hawat S, Knapp B, et al. 2005. J-domain protein CDJ2 and HSP70B are a plastidic chaperone pair that interacts with vesicle-inducing protein in plastids 1. *Mol. Biol. Cell* 16:1165–77
86. Liu Z, Yan H, Wang K, Kuang T, Zhang J, et al. 2004. Crystal structure of spinach major light-harvesting complex at 2.72 Å resolution. *Nature* 428:287–92
87. Lührink J, Samuelsson T, de Gier JW. 2001. YidC/Oxa1p/Alb3: evolutionarily conserved mediators of membrane protein assembly. *FEBS Lett.* 501:1–5
88. Lunde C, Jensen PE, Haldrup A, Knoetzel J, Scheller HV. 2000. The PSI-H subunit of photosystem I is essential for state transitions in plant photosynthesis. *Nature* 408:613–15
89. Maxwell DP, Laudénbach DE, Huner N. 1995. Redox regulation of light-harvesting complex II and cab mRNA abundance in *Dunaliella salina*. *Plant Physiol.* 109:787–95
90. Millar AJ, Straume M, Chory J, Chua NH, Kay SA. 1995. The regulation of circadian period by photo-transduction pathways in *Arabidopsis*. *Science* 267:1163–66
91. Miloslavina Y, Wehner A, Lambrev PH, Wientjes E, Reus M, et al. 2008. Far-red fluorescence: a direct spectroscopic marker for LHCII oligomer formation in non-photochemical quenching. *FEBS Lett.* 582:3625–31
92. Minagawa J, Takahashi Y. 2004. Structure, function and assembly of photosystem II and its light-harvesting proteins. *Photosynth. Res.* 82:241–63
93. Mitra M, Kirst H, Dewez D, Melis A. 2012. Modulation of the light-harvesting chlorophyll antenna size in *Chlamydomonas reinhardtii* by *TLA1* gene over-expression and RNA interference. *Philos. Trans. R. Soc. B* 367:3430–43
94. Mitra M, Melis A. 2010. Genetic and biochemical analysis of the *TLA1* gene in *Chlamydomonas reinhardtii*. *Planta* 231:729–40
95. Moore M, Harrison MS, Peterson EC, Henry R. 2000. Chloroplast Oxa1p homolog albino3 is required for post-translational integration of the light harvesting chlorophyll-binding protein into thylakoid membranes. *J. Biol. Chem.* 275:1529–32
96. Moss DA, Bendall DS. 1984. Cyclic electron transport in chloroplasts: the Q-cycle and the site of action of anthocyanin. *Biochim. Biophys. Acta* 767:389–95
97. Murata N. 1969. Control of excitation transfer in photosynthesis. I. Light-induced change of chlorophyll a fluorescence in *Porphyridium cruentum*. *Biochim. Biophys. Acta* 172:242–51
98. Murchie EH, Niyogi KK. 2011. Manipulation of photoprotection to improve plant photosynthesis. *Plant Physiol.* 155:86–92
99. Mussgnug JH, Wobbe L, Elles I, Claus C, Hamilton M, et al. 2005. NAB1 is an RNA binding protein involved in the light-regulated differential expression of the light-harvesting antenna of *Chlamydomonas reinhardtii*. *Plant Cell* 17:3409–21
100. Nield J, Kruse O, Ruprecht J, da Fonseca P, Buchel C, Barber J. 2000. Three-dimensional structure of *Chlamydomonas reinhardtii* and *Synechococcus elongatus* photosystem II complexes allows for comparison of their oxygen-evolving complex organization. *J. Biol. Chem.* 275:27940–46
101. Niyogi KK, Björkman O, Grossman AR. 1997. *Chlamydomonas* xanthophyll cycle mutants identified by video imaging of chlorophyll fluorescence quenching. *Plant Cell* 9:1369–80
102. Niyogi KK, Grossman AR, Björkman O. 1998. Arabidopsis mutants define a central role for the xanthophyll cycle in the regulation of photosynthetic energy conversion. *Plant Cell* 10:1121–34
103. Nordhues A, Schottler MA, Unger AK, Geimer S, Schonfelder S, et al. 2012. Evidence for a role of VIPP1 in the structural organization of the photosynthetic apparatus in *Chlamydomonas*. *Plant Cell* 24:637–59
104. Nott A, Jung HS, Koussevitzky S, Chory J. 2006. Plastid-to-nucleus retrograde signaling. *Annu. Rev. Plant Biol.* 57:739–59
105. Nymark M, Valle KC, Brembu T, Hancke K, Winge P, et al. 2009. An integrated analysis of molecular acclimation to high light in the marine diatom *Phaeodactylum tricornutum*. *PLoS ONE* 4:e7743

106. Peers G, Truong TB, Ostendorf E, Busch A, Elrad D, et al. 2009. An ancient light-harvesting protein is critical for the regulation of algal photosynthesis. *Nature* 462:518–21
107. Pesaresi P, Hertle A, Pribil M, Kleine T, Wagner R, et al. 2009. Arabidopsis STN7 kinase provides a link between short- and long-term photosynthetic acclimation. *Plant Cell* 21:2402–23
108. Pribil M, Pesaresi P, Hertle A, Barbato R, Leister D. 2010. Role of plastid protein phosphatase TAP38 in LHCII dephosphorylation and thylakoid electron flow. *PLoS Biol.* 8:e1000288
109. Reiland S, Messerli G, Baerenfaller K, Gerrits B, Endler A, et al. 2009. Large-scale *Arabidopsis* phosphoproteome profiling reveals novel chloroplast kinase substrates and phosphorylation networks. *Plant Physiol.* 150:889–903
110. Ruban AV, Berera R, Iliaia C, van Stokkum IH, Kennis JT, et al. 2007. Identification of a mechanism of photoprotective energy dissipation in higher plants. *Nature* 450:575–78
111. Ruban AV, Johnson MP, Duffy CD. 2012. The photoprotective molecular switch in the photosystem II antenna. *Biochim. Biophys. Acta* 1817:167–81
112. Samol I, Shapiguzov A, Ingelsson B, Fucile G, Crevecoeur M, et al. 2012. Identification of a photosystem II phosphatase involved in light acclimation in *Arabidopsis*. *Plant Cell* 24:2596–609
113. Schuenemann D, Gupta S, Persello-Cartieaux F, Klimyuk VI, Jones JD, et al. 1998. A novel signal recognition particle targets light-harvesting proteins to the thylakoid membranes. *Proc. Natl. Acad. Sci. USA* 95:10312–16
114. Shapiguzov A, Ingelsson B, Samol I, Andres C, Kessler F, et al. 2010. The PPH1 phosphatase is specifically involved in LHCII dephosphorylation and state transitions in *Arabidopsis*. *Proc. Natl. Acad. Sci. USA* 107:4782–87
115. Shikanai T. 2007. Cyclic electron transport around photosystem I: genetic approaches. *Annu. Rev. Plant Biol.* 58:199–217
116. Shimoni E, Rav-Hon O, Ohad I, Brumfeld V, Reich Z. 2005. Three-dimensional organization of higher-plant chloroplast thylakoid membranes revealed by electron tomography. *Plant Cell* 17:2580–86
117. Sokolenko A, Fulgosi H, Gal A, Altschmied L, Ohad I, Herrmann RG. 1995. The 64 kDa polypeptide of spinach may not be the LHCII kinase, but a lumen-located polyphenol oxidase. *FEBS Lett.* 371:176–80
118. Sundberg E, Slagter JG, Fridborg I, Cleary SP, Robinson C, Coupland G. 1997. *ALBINO3*, an *Arabidopsis* nuclear gene essential for chloroplast differentiation, encodes a chloroplast protein that shows homology to proteins present in bacterial membranes and yeast mitochondria. *Plant Cell* 9:717–30
119. Takahashi H, Clowes S, Wollman FA, Vallon O, Rappaport F. 2013. Cyclic electron flow is redox-controlled but independent of state transition. *Nat. Commun.* 4:1954
120. Takahashi H, Iwai M, Takahashi Y, Minagawa J. 2006. Identification of the mobile light-harvesting complex II polypeptides for state transitions in *Chlamydomonas reinhardtii*. *Proc. Natl. Acad. Sci. USA* 103:477–82
121. Tikkanen M, Grieco M, Kangasjärvi S, Aro EM. 2010. Thylakoid protein phosphorylation in higher plant chloroplasts optimizes electron transfer under fluctuating light. *Plant Physiol.* 152:723–35
122. Tokutsu R, Kato N, Bui KH, Ishikawa T, Minagawa J. 2012. Revisiting the supramolecular organization of photosystem II in *Chlamydomonas reinhardtii*. *J. Biol. Chem.* 287:31574–81
123. Vainonen JP, Hansson M, Vener AV. 2005. STN8 protein kinase in *Arabidopsis thaliana* is specific in phosphorylation of photosystem II core proteins. *J. Biol. Chem.* 280:33679–86
124. Vener AV, van Kan PJ, Rich PR, Ohad I, Andersson B. 1997. Plastoquinol at the quinol oxidation site of reduced cytochrome *b₆f* mediates signal transduction between light and protein phosphorylation: thylakoid protein kinase deactivation by a single-turnover flash. *Proc. Natl. Acad. Sci. USA* 94:1585–90
125. Wang Q, Sullivan RW, Kight A, Henry RL, Huang J, et al. 2004. Deletion of the chloroplast-localized *Thylakoid formation1* gene product in *Arabidopsis* leads to deficient thylakoid formation and variegated leaves. *Plant Physiol.* 136:3594–604
126. Wientjes E, Drop B, Kouril R, Boekema EJ, Croce R. 2013. During state 1 to state 2 transition in *Arabidopsis thaliana*, the photosystem II supercomplex gets phosphorylated but does not disassemble. *J. Biol. Chem.* 288:32821–26
127. Wientjes E, van Amerongen H, Croce R. 2013. LHCII is an antenna of both photosystems after long-term acclimation. *Biochim. Biophys. Acta* 1827:420–26

128. Willig A, Shapiguzov A, Goldschmidt-Clermont M, Rochaix JD. 2011. The phosphorylation status of the chloroplast protein kinase STN7 of *Arabidopsis* affects its turnover. *Plant Physiol.* 157:2102–7
129. Wobbe L, Blifernez O, Schwarz C, Mussnug JH, Nickelsen J, Kruse O. 2009. Cysteine modification of a specific repressor protein controls the translational status of nucleus-encoded LHCII mRNAs in *Cblamydomonas*. *Proc. Natl. Acad. Sci. USA* 106:13290–95
130. Wollman FA. 2001. State transitions reveal the dynamics and flexibility of the photosynthetic apparatus. *EMBO J.* 20:3623–30
131. Wunder T, Liu Q, Aseeva E, Bonardi V, Leister D, Pribil M. 2013. Control of STN7 transcript abundance and transient STN7 dimerisation are involved in the regulation of STN7 activity. *Planta* 237:541–58
132. Yamamoto HY, Nakayama TO, Chichester CO. 1962. Studies on the light and dark interconversions of leaf xanthophylls. *Arch. Biochem. Biophys.* 97:168–73
133. Zhu XG, Ort DR, Whitmarsh J, Long SP. 2004. The slow reversibility of photosystem II thermal energy dissipation on transfer from high to low light may cause large losses in carbon gain by crop canopies: a theoretical analysis. *J. Exp. Bot.* 55:1167–75
134. Zito F, Finazzi G, Delosme R, Nitschke W, Picot D, Wollman FA. 1999. The Qo site of cytochrome *b₆f* complexes controls the activation of the LHCII kinase. *EMBO J.* 18:2961–69



Contents

| | |
|--|-----|
| Our Eclectic Adventures in the Slower Eras of Photosynthesis: From New England Down Under to Biosphere 2 and Beyond <i>Barry Osmond</i> | 1 |
| Sucrose Metabolism: Gateway to Diverse Carbon Use and Sugar Signaling <i>Yong-Ling Ruan</i> | 33 |
| The Cell Biology of Cellulose Synthesis <i>Heather E. McFarlane, Anett Döring, and Staffan Persson</i> | 69 |
| Phosphate Nutrition: Improving Low-Phosphate Tolerance in Crops <i>Damar Lizbeth López-Arredondo, Marco Antonio Leyva-González, Sandra Isabel González-Morales, José López-Bucio, and Luis Herrera-Estrella</i> | 95 |
| Iron Cofactor Assembly in Plants <i>Janneke Balk and Theresia A. Schaedler</i> | 125 |
| Cyanogenic Glycosides: Synthesis, Physiology, and Phenotypic Plasticity <i>Roslyn M. Gleadow and Birger Lindberg Møller</i> | 155 |
| Engineering Complex Metabolic Pathways in Plants <i>Gemma Farré, Dieter Blancquaert, Teresa Capell, Dominique Van Der Straeten, Paul Christou, and Changfu Zhu</i> | 187 |
| Triterpene Biosynthesis in Plants <i>Ramesha Thimmappa, Katrin Geisler, Thomas Louveau, Paul O'Maille, and Anne Osbourn</i> | 225 |
| To Gibberellins and Beyond! Surveying the Evolution of (Di)Terpenoid Metabolism <i>Jiachen Zi, Sibongile Mafu, and Reuben J. Peters</i> | 259 |
| Regulation and Dynamics of the Light-Harvesting System <i>Jean-David Rochaix</i> | 287 |
| Gene Expression Regulation in Photomorphogenesis from the Perspective of the Central Dogma <i>Shu-Hsing Wu</i> | 311 |

| | |
|--|-----|
| Light Regulation of Plant Defense <i>Carlos L. Ballaré</i> | 335 |
| Heterotrimeric G Protein–Coupled Signaling in Plants <i>Daisuke Urano and Alan M. Jones</i> | 365 |
| Posttranslationally Modified Small-Peptide Signals in Plants <i>Yoshikatsu Matsubayashi</i> | 385 |
| Pentatricopeptide Repeat Proteins in Plants <i>Alice Barkan and Ian Small</i> | 415 |
| Division and Dynamic Morphology of Plastids <i>Katherine W. Osteryoung and Kevin A. Pyke</i> | 443 |
| The Diversity, Biogenesis, and Activities of Endogenous Silencing Small RNAs in <i>Arabidopsis</i> <i>Nicolas G. Bologna and Olivier Voinnet</i> | 473 |
| The Contributions of Transposable Elements to the Structure, Function, and Evolution of Plant Genomes <i>Jeffrey L. Bennetzen and Hao Wang</i> | 505 |
| Natural Variations and Genome-Wide Association Studies in Crop Plants <i>Xuehui Huang and Bin Han</i> | 531 |
| Molecular Control of Grass Inflorescence Development <i>Dabing Zhang and Zheng Yuan</i> | 553 |
| Male Sterility and Fertility Restoration in Crops <i>Letian Chen and Yao-Guang Liu</i> | 579 |
| Molecular Control of Cell Specification and Cell Differentiation During Procambial Development <i>Kaori Miyashima Furuta, Eva Hellmann, and Ykä Helariutta</i> | 607 |
| Adventitious Roots and Lateral Roots: Similarities and Differences <i>Catherine Bellini, Daniel I. Pacurar, and Irene Perrone</i> | 639 |
| Nonstructural Carbon in Woody Plants <i>Michael C. Dietze, Anna Sala, Mariab S. Carbone, Claudia I. Czimczik,</i> <i>Joshua A. Mantooth, Andrew D. Richardson, and Rodrigo Vargas</i> | 667 |
| Plant Interactions with Multiple Insect Herbivores: From Community to Genes <i>Jeltje M. Stam, Anneke Kroes, Yehua Li, Rieta Gols, Joop J.A. van Loon,</i> <i>Erik H. Poelman, and Marcel Dicke</i> | 689 |
| Genetic Engineering and Breeding of Drought-Resistant Crops <i>Honghong Hu and Lizhong Xiong</i> | 715 |

| | |
|---|-----|
| Plant Molecular Pharming for the Treatment of Chronic and Infectious Diseases <i>Eva Stoger, Rainer Fischer, Maurice Moloney, and Julian K.-C. Ma</i> | 743 |
| Genetically Engineered Crops: From Idea to Product <i>Jose Rafael Prado, Gerrit Segers, Toni Voelker, Dave Carson, Raymond Dobert, Jonathan Phillips, Kevin Cook, Camilo Cornejo, Josh Monken, Laura Grapes, Tracey Reynolds, and Susan Martino-Catt</i> | 769 |

Errata

An online log of corrections to *Annual Review of Plant Biology* articles may be found at
<http://www.annualreviews.org/errata/arplant>



ANNUAL REVIEWS

It's about time. Your time. It's time well spent.

New From Annual Reviews:

Annual Review of Statistics and Its Application

Volume 1 • Online January 2014 • <http://statistics.annualreviews.org>

Editor: **Stephen E. Fienberg**, *Carnegie Mellon University*

Associate Editors: **Nancy Reid**, *University of Toronto*

Stephen M. Stigler, *University of Chicago*

The *Annual Review of Statistics and Its Application* aims to inform statisticians and quantitative methodologists, as well as all scientists and users of statistics about major methodological advances and the computational tools that allow for their implementation. It will include developments in the field of statistics, including theoretical statistical underpinnings of new methodology, as well as developments in specific application domains such as biostatistics and bioinformatics, economics, machine learning, psychology, sociology, and aspects of the physical sciences.

Complimentary online access to the first volume will be available until January 2015.

TABLE OF CONTENTS:

- *What Is Statistics?* Stephen E. Fienberg
- *A Systematic Statistical Approach to Evaluating Evidence from Observational Studies*, David Madigan, Paul E. Stang, Jesse A. Berlin, Martijn Schuemie, J. Marc Overhage, Marc A. Suchard, Bill Dumouchel, Abraham G. Hartzema, Patrick B. Ryan
- *The Role of Statistics in the Discovery of a Higgs Boson*, David A. van Dyk
- *Brain Imaging Analysis*, F. DuBois Bowman
- *Statistics and Climate*, Peter Guttorp
- *Climate Simulators and Climate Projections*, Jonathan Rougier, Michael Goldstein
- *Probabilistic Forecasting*, Tilmann Gneiting, Matthias Katzfuss
- *Bayesian Computational Tools*, Christian P. Robert
- *Bayesian Computation Via Markov Chain Monte Carlo*, Radu V. Craiu, Jeffrey S. Rosenthal
- *Build, Compute, Critique, Repeat: Data Analysis with Latent Variable Models*, David M. Blei
- *Structured Regularizers for High-Dimensional Problems: Statistical and Computational Issues*, Martin J. Wainwright
- *High-Dimensional Statistics with a View Toward Applications in Biology*, Peter Bühlmann, Markus Kalisch, Lukas Meier
- *Next-Generation Statistical Genetics: Modeling, Penalization, and Optimization in High-Dimensional Data*, Kenneth Lange, Jeanette C. Papp, Janet S. Sinsheimer, Eric M. Sobel
- *Breaking Bad: Two Decades of Life-Course Data Analysis in Criminology, Developmental Psychology, and Beyond*, Elena A. Erosheva, Ross L. Matsueda, Donatello Telesca
- *Event History Analysis*, Niels Keiding
- *Statistical Evaluation of Forensic DNA Profile Evidence*, Christopher D. Steele, David J. Balding
- *Using League Table Rankings in Public Policy Formation: Statistical Issues*, Harvey Goldstein
- *Statistical Ecology*, Ruth King
- *Estimating the Number of Species in Microbial Diversity Studies*, John Bunge, Amy Willis, Fiona Walsh
- *Dynamic Treatment Regimes*, Bibhas Chakraborty, Susan A. Murphy
- *Statistics and Related Topics in Single-Molecule Biophysics*, Hong Qian, S.C. Kou
- *Statistics and Quantitative Risk Management for Banking and Insurance*, Paul Embrechts, Marius Hofert

Access this and all other Annual Reviews journals via your institution at www.annualreviews.org.

ANNUAL REVIEWS | Connect With Our Experts

Tel: 800.523.8635 (US/CAN) | Tel: 650.493.4400 | Fax: 650.424.0910 | Email: service@annualreviews.org

