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Plants and algae have versatile abilities to acclimate themselves to changing environments. We are interested in these acclimation processes, and how they efficiently yet safely harness sunlight for photosynthesis under fluctuating light conditions. Using a model green alga, we are studying the molecular mechanisms underlying photoacclimation of photosynthetic machinery. We are also applying knowledge obtained in the studies of this model green alga to various photosynthetic organisms, including phytoplankton and vascular plants, to explore how environmentally important photosynthetic organisms thrive in their ecological niche.



Visual overview of this lab's work.

I. Structural characterization of the photosystems in the green alga *Chlorella sorokiniana*.

The photosynthetic conversion of light energy into chemical energy is performed by photosystems II and I (PSII and PSI) embedded within the thylakoid membranes. In plants and green algae, PSII and PSI comprise the core complex and light-harvesting complexes (LHCII and LHCI), forming PSII-LHCII and PSI-LHCI supercomplexes, respectively. The structural information about photosystem supercomplexes of green algae has been limited to chlorophytic algae. Here, to obtain an insight into the evolution of Chlorophyta, we determined the supramolecular organization of the PSII-LHCII and PSI-LHCI supercomplexes from the freshwater green alga *Chlorella sorokiniana*, which belongs to Trebouxiophyceae (Fig. 1) (Watanabe and Minagawa, *Planta*, 252:79). The obtained results showed that the supramolecular organizations of the photosystem supercomplexes in *C. sorokiniana* were essentially the same as those of the model green alga *C. reinhardtii*, which belongs to Chlorophyceae, namely PSII-LHCII supercomplex formed the C₂S₂M₂L₂ configuration and PSI-LHCI supercomplex was associated with 10 LHCI subunits.

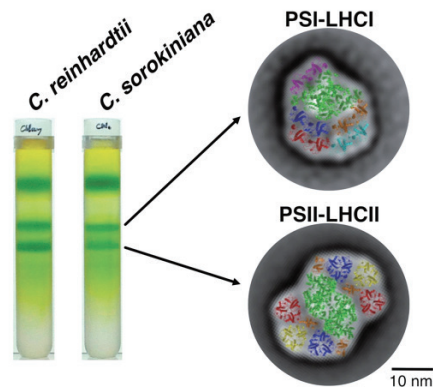


Figure 1. Single-particle EM analysis of photosystems isolated from SDG ultracentrifugation of the α -DDM-solubilized thylakoids from *C. sorokiniana* cells. Watanabe and Minagawa (2020) *Planta*, 252:79.

II. Characterization of a giant photosystem I supercomplex in the symbiotic dinoflagellate Symbiodiniaceae.

Symbiodiniaceae are symbiotic dinoflagellates that provide photosynthetic products to corals. Because corals are distributed across a wide range of depths in the ocean, Symbiodiniaceae species must adapt to various light environments to optimize their photosynthetic performance. However, as few biochemical studies of Symbiodiniaceae photosystems have been reported, the molecular mechanisms of photoadaptation in this algal family remain poorly understood. Here, to investigate the photosynthetic machineries in Symbiodiniaceae, we purified and characterized the PSI supercomplex from the genome-sequenced *Breviolum minutum* (formerly *Symbiodinium minutum*) (Fig. 2) (Kato *et al.*, *Plant Physiol.*, 183:1725-1734). Mass spectrometry analysis revealed 25 light-harvesting complexes (LHCs), including both LHCF and LHCR families, from the purified PSI-LHC supercomplex. Single-particle electron microscopy

showed unique giant supercomplex structures of PSI that were associated with the LHCs. Moreover, the PSI-LHC supercomplex contained a significant amount of the xanthophyll cycle pigment diadinoxanthin. Upon high light treatment, *B. minutum* cells showed increased nonphotochemical quenching, which was correlated with the conversion of diadinoxanthin to diatoxanthin, occurring preferentially in the PSI-LHC supercomplex. The possible role of PSI-LHC in photoprotection in Symbiodiniaceae was discussed.

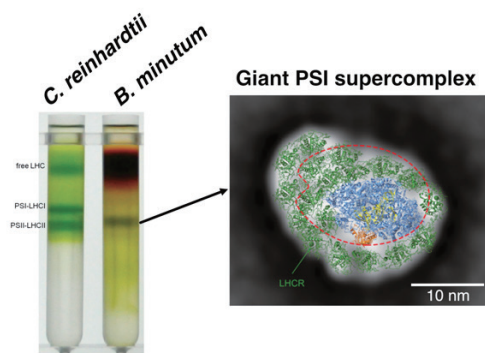


Figure 2. Single-particle EM analysis of photosystem I isolated from SDG ultracentrifugation of the α -DDM-solubilized thylakoids from *B. minutum* cells. Kato *et al.* (2020) *Plant Physiol.*, 183:1725-1734.

III. Multimeric and monomeric photosystem II supercomplexes represent structural adaptations to low- and high-light conditions.

An intriguing molecular architecture called the “semi-crystalline photosystem II (PSII) array” has been observed in the thylakoid membranes in vascular plants. It is an array of PSII–LHCII supercomplexes that only appears in low light, but its functional role has not been clarified. Here, we identified PSII–LHCII supercomplexes in their monomeric and multimeric forms in low light–acclimated spinach leaves and prepared them using sucrose-density gradient ultracentrifugation in the presence of amphipol A8-35. When the leaves were acclimated to high light, only the monomeric forms were present, suggesting that the multimeric forms represent a structural adaptation to low light and that disaggregation of the PSII–LHCII supercomplex represents an adaptation to high light. Single-particle EM revealed that the multimeric PSII–LHCII supercomplexes are composed of two (“mega-complex”) or three (“arraycomplex”) units of PSII–LHCII supercomplexes, which likely constitute a fraction of the semi-crystalline PSII array. Further characterization with fluorescence analysis revealed that multimeric forms have a higher light-harvesting capability but a lower thermal dissipation capability than the monomeric form. These findings suggest that the configurational conversion of PSII–LHCII supercomplexes may serve as a structural basis for acclimation of plants to environmental light (Fig. 3) (Kim and Watanabe *et al.*, *J. Biol. Chem.*, 295:14537-14545).

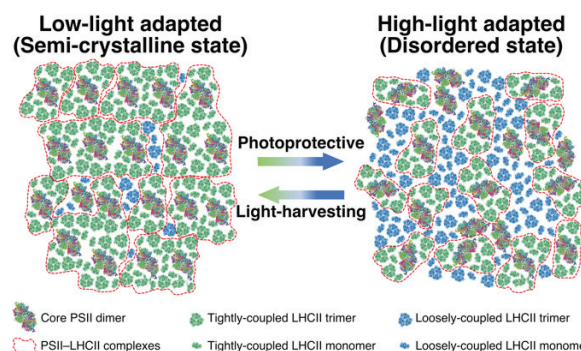


Figure 3. Schematic representation of the putative arrangements of PSII–LHCII supercomplexes in the thylakoid membrane in low-light and high-light conditions. Kim, Watanabe *et al.* (2020) *J. Biol. Chem.*, 295:14537-14545.

IV. Photoprotective capabilities of light-harvesting complex II trimers in a green alga *Chlamydomonas reinhardtii*.

Major light-harvesting complex (LHCII) trimers in plants induce the thermal dissipation of absorbed excitation energy against photooxidative damage under excess light conditions. LHCII trimers in green algae have been thought to be incapable of energy dissipation without additional quencher proteins, although LHCII in plants and green algae are homologous. In this study, we investigated the energy-dissipative capabilities of four distinct types of LHCII trimers isolated from the model green alga *Chlamydomonas reinhardtii* using spectroscopic analysis. Our results revealed that the LHCII trimers possessing LHCII type II (LHCBM5) and LHCII type IV (LHCBM1) had efficient energy-dissipative capabilities, whereas LHCII type I (LHCBM3/4/6/8/9) and type III (LHCBM2/7) did not. On the basis of the amino acid sequences of LHCBM5 and LHCBM1 compared with the other LHCBMs, we propose that positively charged extra N-terminal amino acid residues mediate the interactions between LHCII trimers to form energy-dissipative states (Fig. 4) (Kim *et al.*, *J. Phys. Chem. Lett.*, 11:7755-7761).

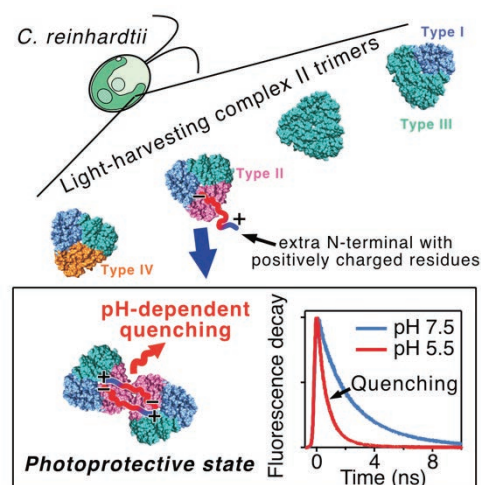


Figure 4. Schematic representation of the extra N-terminal-dependent photoprotective states of LHCII trimers from *C. reinhardtii*. Kim *et al.* (2020) *J. Phys. Chem. Lett.*, 11:7755-7761.

Publication List:

[Original Papers]

- Faktorova, D., Nisbet, R.E.R., Robledo, J.A.F., Casacuberta, E., Sudek, L., Allen, A.E., Ares Jr., M., Areste, C., Balestreri, C., Barbrook, A.C., *et al.* (2020). Genetic tool development in marine protists: emerging model organisms for experimental cell biology. *Nat. Methods* 17, 481–494. DOI: 10.1038/s41592-020-0796-x
- Kato, H., Tokutsu, R., Kubota-Kawai, H., Burton-Smith, R.N., Kim, E., and Minagawa, J. (2020). Characterization of a giant PSI Supercomplex in the symbiotic dinoflagellate symbiodiniaceae. *Plant Physiol.* 183, 1725–1734. DOI: 10.1104/pp.20.00726
- Kim, E., Kawakami, K., Sato, R., and Ishii A., and Minagawa, J. (2020). Photoprotective capabilities of light-harvesting complex II trimers in the green alga *Chlamydomonas reinhardtii*. *J. Phys. Chem. Lett.* 11, 7755–7761. DOI: 10.1021/acs.jpcllett.0c02098
- Kim, E., Watanabe, A., Duffy, C.D.P., Ruban, A. V, and Minagawa, J. (2020). Multimeric and monomeric photosystem II supercomplexes represent structural adaptations to low- and high-light conditions. *J. Biol. Chem.* 295, 14537–14545. DOI: 10.1074/jbc.RA120.014198
- Kishimoto, M., Baird, A.H., Maruyama, S., Minagawa, J., and Takahashi, S. (2020). Loss of symbiont infectivity following thermal stress can be a factor limiting recovery from bleaching in cnidarians. *ISME J.* 14, 3149–3152. DOI: 10.1038/s41396-020-00742-8
- Shukla, M.K., Watanabe, A., Wilson, S., Giovagnetti, V., Moustafa, E.I., Minagawa, J., and Ruban, V. A. (2020). A novel method produces native light-harvesting complex II aggregates from the photosynthetic membrane revealing their role in nonphotochemical quenching. *J. Biol. Chem.* 295, 17816–17826. DOI: 10.1074/jbc.RA120.016181
- Tokutsu, R., Fujimura-Kamada, K., Yamasaki, T., Okajima, K., and Minagawa, J. (2021). UV-A/B radiation rapidly activates photoprotective mechanisms in *Chlamydomonas reinhardtii*. *Plant Physiol.* 185, 1894–1902. DOI: 10.1093/plphys/kiab004
- Watanabe, A., and Minagawa, J. (2020). Structural characterization of the photosystems in the green alga *Chlorella sorokiniana*. *Planta* 252, 79. DOI: 10.1007/s00425-020-03487-y