pigment–protein complexes that occur asynchronously and therefore cannot be resolved in an ensemble average of typical bulk measurements.

By observing single-protein complexes instead, Schlau-Cohen and co-workers have now demonstrated that there is actually more than one dissipating state in the light-harvesting complex stress-related (LHCSR) protein of algae, thereby resolving an important part in this discussion. LHCSR has been recently discovered to be essential for the regulation of light-harvesting in green algae[7,8] that make up a major part of photosynthetic energy conversion on Earth[9]. To resolve the molecular transformations causing dissipative channels, the team investigated hundreds of individual complexes of LHCSR, as well as normal light-harvesting pigment–protein complexes and compared their behaviour in the absence and presence of zeaxanthin or violaxanthin. They also studied different pH values typically observed under different light conditions.

The actual regulation status and light responsiveness of photosynthetic organisms is usually monitored by the residual intensity of chlorophyll fluorescence, as the same mechanisms that dissipate excess energy also intrinsically reduce the energy available for residual fluorescence[10]. Thus, Schlau-Cohen and co-workers also measured the same parameter but monitored single light-harvesting complexes instead. To do this they attached the complexes to cover slips in an aqueous buffer environment and observed them in a fluorescence microscope with pulsed excitation. This allowed the team to resolve the fluorescence lifetime of chlorophyll in individual complexes in addition to the intensity. In contrast to measurements using entire photosynthetic organisms, in which fluorescence parameters change gradually in response to varying light conditions, the observation of individual pigment–protein complexes displayed drastic reversible or non-reversible jumps in both parameters. These correspond to different states that the complexes adopted when irradiated with light intensities that photosynthetic organisms typically experience during a day. The group also looked at the magnitudes and timescales of the fluctuations in the fluorescence parameters and analysed a large number of such time traces with an algorithm that identified the number of different states in the data and their overall occurrence. With LHCSR they observed quite distinct states with very different energy dissipation characteristics that heavily depended on the carotenoid composition and pH values. While low pH values strongly shifted the equilibrium to states that have significantly lower fluorescence intensities, the presence of Zea shifted the equilibrium to distinct states that have additionally much shorter fluorescence lifetimes. If low pH as well as Zea were present, both worked together to result in the smallest intensities and lifetimes. These data demonstrate that LHCSR can exist in at least three states — one non-dissipative and two-to-three dissipative — that depend on the pH values as well as carotenoid composition.

Based on these observations the model proposed by Schlau-Cohen and co-workers is very straightforward. A reversible change in the pH-gradient is a very quick response as a function of excess light, and can explain the regulation observed on a timescale of a few seconds when the intensity of light fluctuates wildly (Fig. 1a,b). Changes on longer timescales enable reversible interconversion of violaxanthin to zeaxanthin, and vice versa, on a timescale of minutes to hours thereby opening an additional channel of energy dissipation (Fig. 1a,c). Both together allow for full switching from efficient light harvesting and energy conversion of each available photon (Fig. 1a) to high dissipation of excess energy (Fig. 1d) on different timescales while maintaining an optimum flux of energy towards the reaction centre.

One might argue that the measurement of single-protein complexes at different pH values does not reflect physiological conditions as the complexes are not embedded in native membranes and also because under physiological conditions there is a transmembrane pH gradient present rather than a homogenously applied single pH. Such arguments have to be carefully kept in mind when extending conclusions obtained from isolated, single-protein complexes to the situation *in vivo*. However, the data of Schlau-Cohen and co-workers clearly demonstrate that LHCSR can adopt at least three different states that correlate directly to the presence and absence of Zea, as well as low pH values, and that the induced states have energy dissipative character as observed *in vivo*. It is likely that these states are at least partly caused by different conformations of the complexes.

The data do not yet show to what extent different photophysical quenching mechanisms are contributing to the observed dissipating character in isolated complexes as well as *in vivo*, and what molecular mechanisms, such as conformational transitions, are involved. However, to understand the whole, one must also understand the properties of the individual parts, and the work presented by Schlau-Cohen and co-workers has laid an excellent basis to further study these exciting questions in the important field of bioenergetics. ❐

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Correction

A sentence in the fifth paragraph of the In Your Element article 'V for vanadium' (*Nat. Chem.* **9,** 602; 2017) incorrectly mentioned magnesium, it should have read: "Their rich redox chemistry is also key to their application in biological systems (think manganese in photosynthesis)". This has been corrected after print 21 June 2017.