

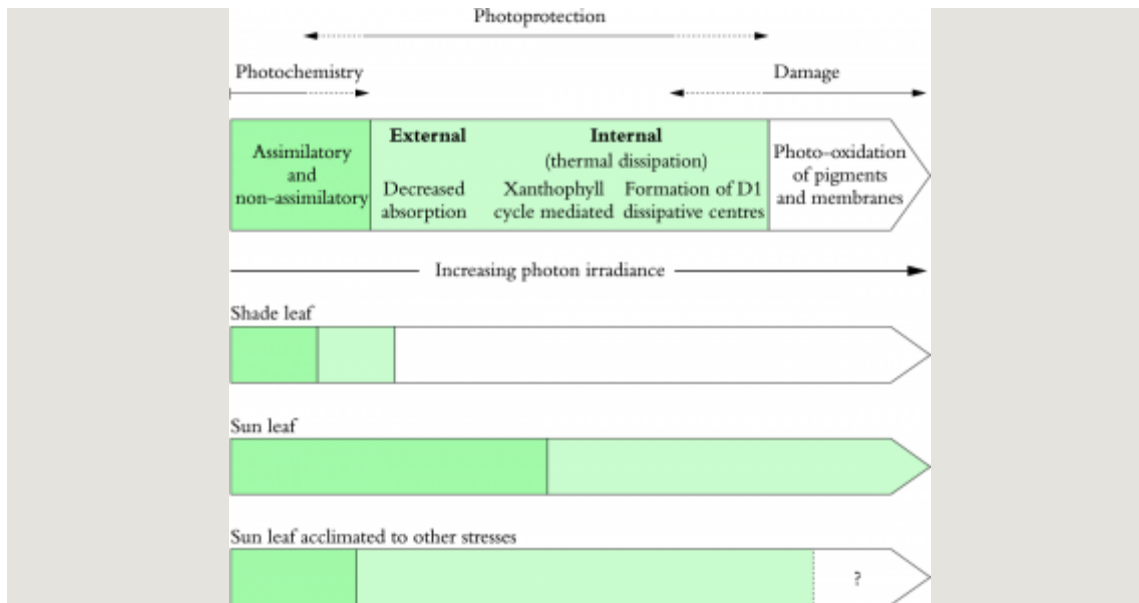
Photoinhibition and photoprotection

In light-response curves for photosynthesis (Figure 12.7), photosynthesis is light limited, as implied by the initial linear region of the curve. However, once a light-saturation point has been reached, further light exceeds the energy-utilising capacity of that photosynthesising leaf. Dashed lines in Figure 12.7 represent a continuation of the initial rate of light processing (quantum yield of photosynthesis) and show actual light absorption. The extent to which this absorbed light is not 'gainfully employed' for photosynthesis is set by P_{\max} (light-saturated rate of photosynthesis in normal air). At low light ($< 100\mu\text{mol quanta m}^{-2}\text{ s}^{-1}$), both sun and shade leaves use more than 80% of absorbed light for photosynthesis.

Once P_{\max} has been reached, all additional light is in excess, and since shade plants have a lower P_{\max} than sun plants, they experience more excess light at a given photon irradiance above saturation. Additional stresses such as drought, nutrient limitation or temperature extremes can lead to a reduction in P_{\max} and thus increase the probability that plants will be exposed to excess light. However, even the most hardy sun plant will reach P_{\max} at less than full sunlight (incident beam normal to leaf surface). At that level (say, $1000\mu\text{mol quanta m}^{-2}\text{ s}^{-1}$) approximately 25% of absorbed energy is used in driving photosynthesis, but at full sunlight (c. $2000\mu\text{mol quanta m}^{-2}\text{ s}^{-1}$) as little as 10% is used in this way (Long *et al.* 1994). Individual leaves on plants growing in full sun commonly experience excess light.

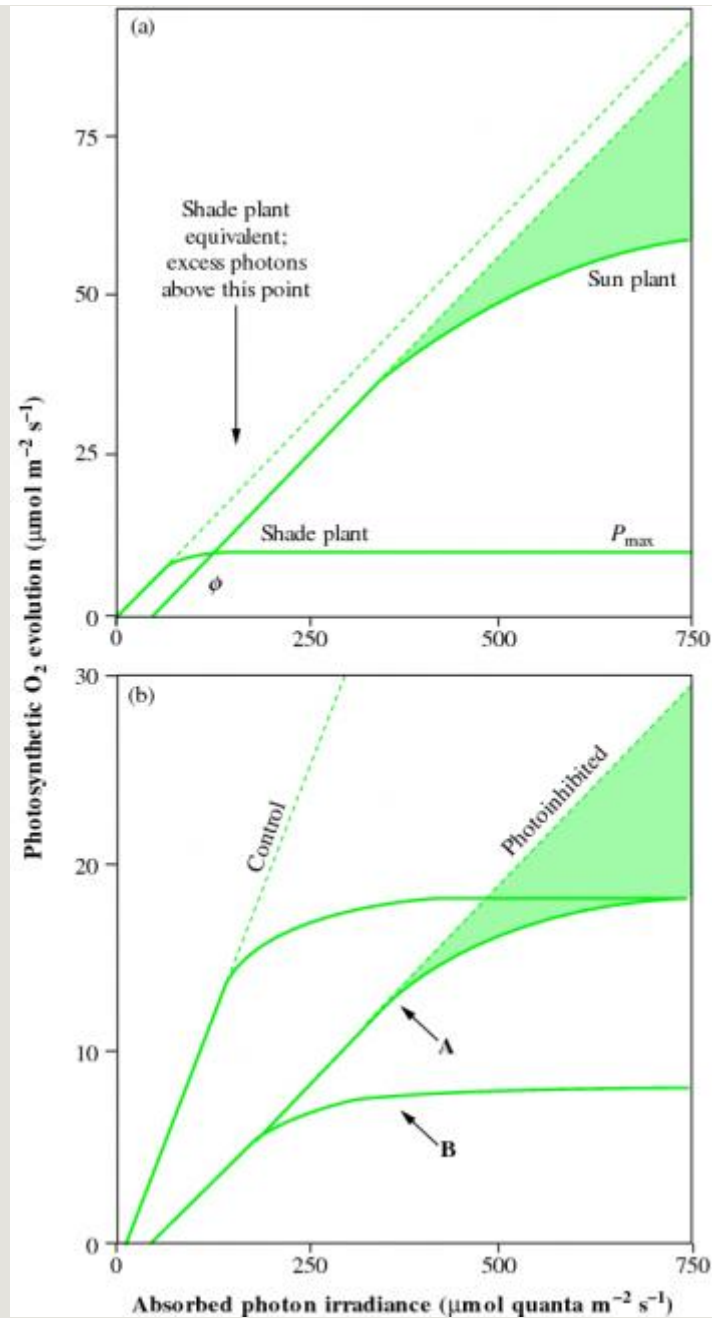
Such light is potentially damaging, and plants adapted to full sunlight have evolved with a number of mechanisms for either avoiding excess light or for dissipating excess absorbed energy.

Mechanisms for avoiding high light such as leaf angle and surface features, forestall absorption of excess light. Rapid responses, such as changes in leaf angle in *Oxalis* and *Omalanthus*, occur in a matter of minutes and can regulate light interception on a diurnal basis. Slower-acting mechanisms including production of wax on leaves will be useful where there has been a sustained change in light environment. These kinds of mechanisms constitute *external* photoprotection.



As photon irradiance is increased, energy utilisation gives way progressively to energy dissipation. Photosynthetic events shift from photochemistry, to external and then internal photoprotection and finally photodamage. These processes are interconnected and show considerable overlap. Their comparative importance for shade leaves and sun leaves is indicated. Photoprotection is especially well expressed in sun leaves acclimated to additional environmental and biotic stresses

(Based on Demmig-Adams and Adams 1992; reproduced with permission of the *Annual Review of Plant Physiology and Plant Molecular Biology*)

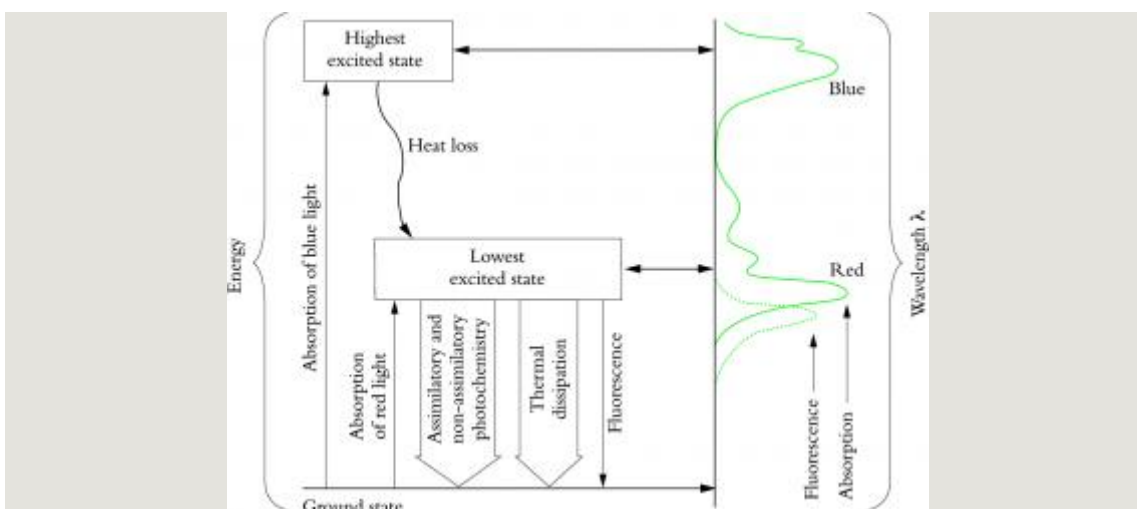


Photosynthesis and photoinhibition in sun and shade leaves. (a) shows photosynthesis-light response curves for sun and shade plants as in Figure 12.7. Dashed lines represent light absorbed by each plant. Shaded areas represent absorption of excess energy that has to be dissipated if the sun plant is to avoid damage. Shade plants have an even greater need because they absorb more light (more chlorophyll per unit leaf mass), but need less light to saturate photosynthesis. (b) shows (left side) a non-inhibited control shade leaf corresponding to that in (a) and (right side) the result of prolonged exposure to excess light (note scale expansion on the ordinate of (b)). In curve A, photoprotective processes have reduced the quantum yield of photosynthesis, but P_{max} is unchanged. By implication, more light is required to produce the same amount of O₂ (quantum yield is lower) but P_{max} can be sustained by providing more light. Further exposure to excess light results in curve B where both quantum yield and P_{max} are reduced. Photosynthetic rate is then reduced at all light levels, and recover is slower compared to leaves in curve A

(Based on Osmond 1994, with acknowledgement to Bios Scientific Publishers)

Absorption of excess sunlight often leads to photoinhibition, defined here as a light-dependent downregulation of the quantum yield of photosynthesis (Figure 12.9). Ironic as it might seem, photoinhibition is one of the most important regulatory mechanisms in photosynthesis, and results from a series of internal photoprotective mechanisms which act to reduce the amount of light reaching the reaction centres of PSII. One immediate consequence is a reduction in quantum yield (expressed in terms of *absorbed* light).

Photoprotection (Figure 12.8) is normally sufficient to cope with light absorbed by leaves; photodamage only occurs when the capacity for photoprotection is exhausted. Photodamage is manifest as a decline in both quantum yield and P_{max} , and recovers only very slowly (hours to days), whereas photoinhibition recovers much faster (minutes to hours). Severe photodamage results in bleaching of pigments and damage to membranes (photo-oxidation) and may lead to tissue death.



Absorption of blue or red light (wavebands corresponding to leaf absorption maxima; Figure 1.3) leads to excitation of chlorophyll from its ground state. Although blue light has higher energy and causes excitation to a higher excited state, this energy is quickly lost as heat, and chlorophyll molecules drop to a lower excited state. Utilisation of energy from excited chlorophyll molecules results in either assimilatory or non-assimilatory photochemistry, thermal dissipation or release of light of a longer wavelength (fluorescence). Arrow thickness indicateds comparative importance of each process for energy utilization.

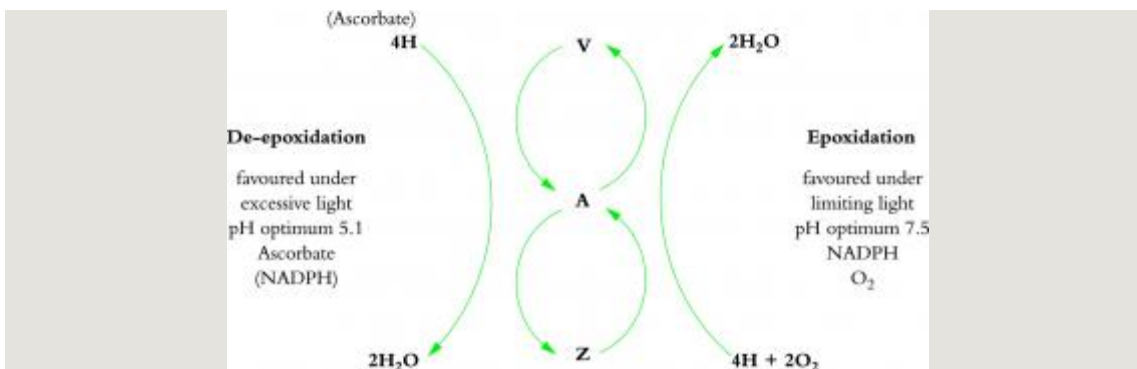
(Based on Taiz and Seiger 1991)

Consider the alternative fates of light energy absorbed by a leaf and their relevance to photoprotection, photoinhibition and photodamage. The proportion of absorbed energy consumed by these different sequences dictates their comparative significance, and in order of importance as protective devices they are:

1. assimilatory photochemistry, leading to fixation of CO₂ into stable chemical products;
2. non-assimilatory photochemistry, that is, energy consumption by metabolic processes that do not result in fixation of CO₂ into stable chemical products such as photorespiration, nitrate reduction and the Mehler reaction. All consume energy but there is no net gain in carbon as a result;
3. conversion of light energy into heat (thermal dissipation);

4. re-emission of photons as fluorescence. Emission of *in vivo* Chl *a* fluorescence is revealed dramatically during measurements. Such emission still accounts for only *c.* 1% of energy derived from absorbed light.

Most of the NADPH and ATP formed during photosynthetic energy transduction is stored as stable photosynthetic products. Some is consumed in photorespiration and nitrate reduction. Because these non-assimilatory processes also utilise NADPH and ATP they help reduce the need for photoprotection. The Mehler reaction, in which electrons flow to O₂ via photosynthetic system I (PSI) (Figure 1.10), still supports electron flow and thus might also reduce a need for photoprotection (Osmond and Grace 1995).



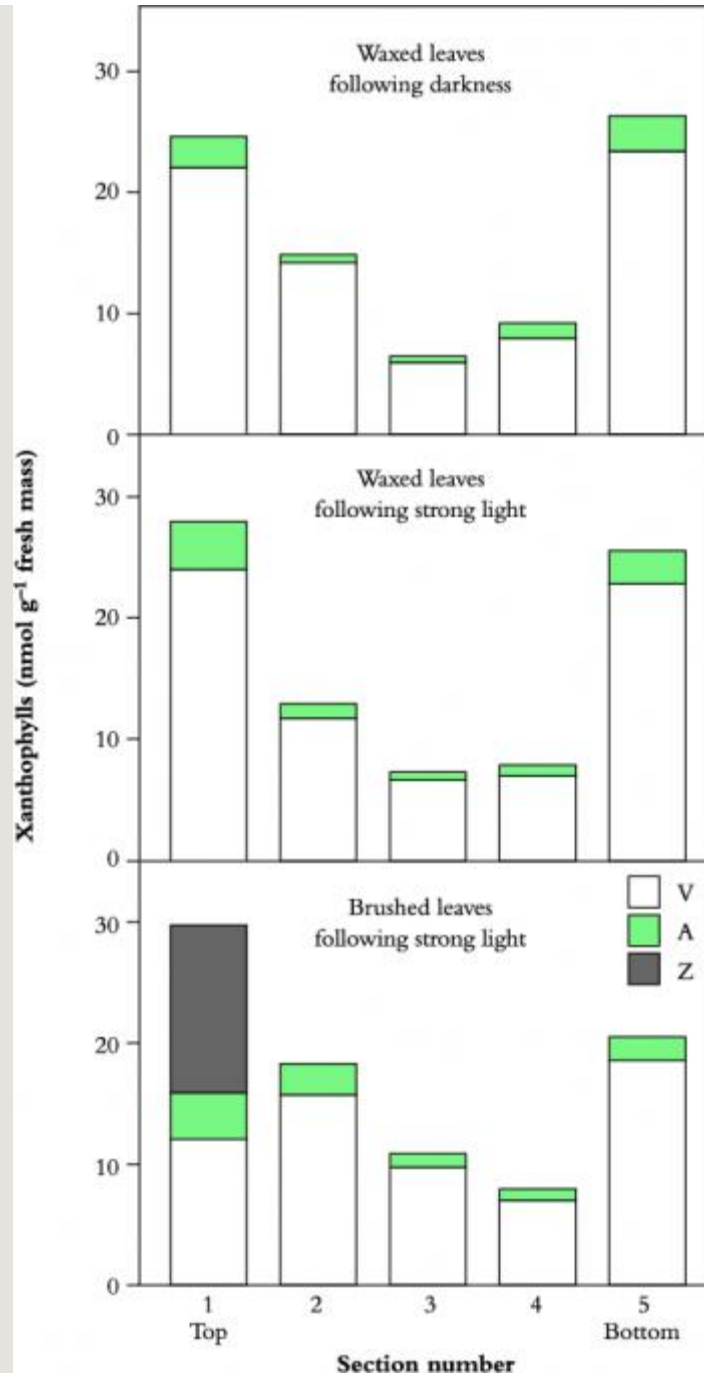
The xanthophyll cycle summarised here contributes to dissipation of excess light energy, and involves three pigments, violaxanthin (V), antheraxanthin (A) and zeaxanthin (Z). All occur in chloroplast thylakoid membranes. This special group of carotenoids undergoes interconversion in response to high light. In low light, violaxanthin predominates but excess light levels produce chloroplast conditions which are conducive to de-epoxidation of violaxanthin to zeaxanthin (conditions summarised on the left hand side). Zeaxanthin, and possibly antheraxanthin, provide photoprotective thermal dissipation of excess light energy. When light levels are no longer excessive, chloroplast conditions favour epoxidation of zeaxanthin back to violaxanthin (summarised on the right-hand side).

(Original drawing courtesy S.A. Robinson)

However, if photochemical capacity is exceeded by incoming energy, a plant will engage photoprotective mechanisms which increase the amount of energy dissipated as heat. This non-photochemical conversion of light energy is thought to occur in the PSII antennae and involves a group of pigments known as xanthophylls and including violaxanthin, antheraxanthin and zeaxanthin (Figure 12.11). These are a special group of carotenoids which undergo interconversion in response to excess light. Energy is dissipated in the process. In low light, violaxanthin predominates, but when light is in excess, conversion to zeaxanthin via antheraxanthin occurs. This conversion requires a low pH, ascorbate and NADPH.

Such conditions exist in the lumen of chloroplasts in high light. When light levels are no longer excessive zeaxanthin slowly converts back to violaxanthin via antheraxanthin. Total pool sizes of the xanthophyll pigments increase with increasing exposure to excess light. Sun plants can have three- to four-fold larger pools of violaxanthin, antheraxanthin and zeaxanthin than shade plants and the presence of other stresses can also result in increases in pool size.

Internal differences between sun and shade leaves with respect to energy dissipation are also apparent in different patterns of attenuation of light through mesophyll tissues. This is nicely illustrated by *Cotyledon orbiculata*, the CAM plant cited earlier. Here, xanthophylls are mostly found in outermost cell layers where the light environment is strongest. If the reflective wax coating is intact no internal photoprotection is required at growth irradiance and there is no zeaxanthin formed. However, if surface wax (external photo-protection) is removed by hand, internal photoprotection is then needed and zeaxanthin appears in the outermost layer.



External and internal photoprotection in thick leaves of *Cotyledon orbiculata*. Removal of protective wax from upper surfaces stimulates synthesis of zeaxanthin. Overall activity of the xanthophyll cycle varies with mesophyll irradiance, while concentration of particular components also varies according to tissue depth. Changes in concentration of the

various components of the xanthophyll cycle are shown here for successive (1 mm) layers in thick leaves of *C. orbiculata*, with and without their wax coating. In waxed leaves at the end of a dark period (top figure) xanthophyll cycle pigments are represented mainly by violaxanthin (V) with a small amount of antheraxanthin (A). After 6 h exposure to high light (middle figure) a small amount of violaxanthin is converted to antheraxanthin, but no zeaxanthin (Z) is formed, indicating that the natural wax coating on these leaves is protecting chloroplasts from excess light. Physical removal of surface wax (brushed leaves in bottom figure) results in zeaxanthin production within the uppermost layer of leaf tissue but not in deeper tissues or lower surfaces. Restriction of this xanthophyll cycle component to the top section confirms that zeaxanthin accumulation is a response to excess light

(Based on Robinson and Osmond 1994)