Plants, like most other organisms, require sunlight to thrive. But exposure to sunlight has its perils, and these appear to be on the increase: the level of harmful solar UV radiation reaching the earth’s surface is rising as ozone levels decline. Plants exposed to excess UV radiation show alterations in photosynthesis, growth and morphology, flavonoid production, and a host of other physiological processes (for reviews, see Stapleton, 1992; Bornman and Teramura, 1993). UV radiation directly alters the structure of DNA as well as damaging proteins or membranes or harming nucleic acids indirectly (e.g., by cross-linking them to proteins). Two major products of direct DNA damage are dimers of adjacent pyrimidines: cyclobutane pyrimidine dimers (CPDs) and pyrimidine(6-4)pyrimidone dimers [(6-4) photoproducts]. UV radiation can also generate single-strand breaks.

All organisms that are exposed to sunlight, and even some that are not, have mechanisms to protect against UV damage. For example, UV-absorbing molecules in human skin (melanin), in plant epidermis (flavonoids), and in the sheath of blue-green algae (mycosporine-like amino acid compounds) are thought to filter out some of the harmful wavelengths, reducing the amount of UV that penetrates the organism. A direct demonstration of the importance of flavonoids in protecting plants from the damaging effects of UV radiation was recently provided by an analysis of the Arabidopsis mutants tfl4 and tfl5 (Li et al., 1993). These mutants, which are defective in flavonoid biosynthesis, are much more sensitive to UV irradiation than are wild-type plants. Many organisms have also evolved mechanisms in which UV radiation actually induces the accumulation of protective molecules; in plants, for instance, flavonoids are induced upon UV-B exposure (Beggs and Wellman, 1985). The details of how UV is perceived by the cells and how this perception is transduced to affect gene expression have yet to be worked out in any system.

DNA damage by UV radiation is countered not only by the presence of protective molecules but also by repair systems. Genetic, molecular, and biochemical studies of Escherichia coli, yeast, and mammalian cell mutants hypersensitive to DNA damaging agents, including UV radiation, have shown that UV-induced DNA damage can be repaired by several distinct mechanisms. One mechanism is photoreactivation, in which the enzyme photolyase, in the presence of light, cleaves CPDs and returns the bases to their original state. The other class of pyrimidine dimers, (6-4) photoproducts (as well as CPDs in the dark or in the absence of photolyase), are repaired by an excision repair process in which incisions are made on either side of the lesion, an oligonucleotide stretch containing the lesion is removed, and a replacement strand is synthesized. Recent evidence indicates that excision repair of (6-4) photoproducts may in some cases be enhanced by photoreactivating wavelengths (Mitchell and Karentz, 1993), but only in Drosophila is there any evidence that (6-4) photoproducts can actually be photoreactivated (Todo et al., 1993). Both CPDs and (6-4) photoproducts, as well as DNA strand breaks, can also be repaired during or after replication, either by recombinational repair or by error-prone repair, in which polymerase inserts any base opposite a base so damaged as to be unrecognizable.

At least some of these repair mechanisms operate in plants. For example, Pang and Hays (1991) demonstrated that Arabidopsis extracts contain UV-inducible photolyase activity, and a photolyase gene whose transcript accumulates with exposure to white light has been cloned from mustard (Batschauer, 1993). Photolyase activity has also been detected in maize and bean, and photoreactivation has been observed in several other plant species (for review, see McClennan, 1987). In addition, evidence of plant excision repair has come from studies of grass pea and of carrot protoplasts (McClennan, 1987). As yet, however, postreplicative repair has not been demonstrated in plants.

Although biochemical studies have begun to shed light on the biochemistry of plant DNA repair, to learn about the full range of UV sensing and protection mechanisms plants have at their disposal and to identify potentially novel plant-specific repair mechanisms, it is necessary to study mutants in which sensitivity to UV radiation is increased or decreased relative to that of the wild type. Working with Arabidopsis, Britt et al. (1993) recently identified such a UV-hypersensitive mutant, dubbed uvr7, and in this issue, Harlow and coworkers (pages 227-235) describe the identification and characterization of a second UV-hypersensitive mutant of Arabidopsis, uvr7. In both mutants, the UV hypersensitivity appears to arise from a defect in DNA repair.

To isolate UV-hypersensitive mutants, Britt et al. (1993) screened for seedlings whose root growth was halted by an acute dose of UV-B that is tolerated by progenitor seedlings. Harlow and coworkers used a different approach, screening rosette plants for mutants whose leaves were visibly damaged by an acute dose of UV-C that produces no visible damage to the leaves of wild-type plants. Although the uvr7 mutant was identified by its increased sensitivity to UV-C, it is also hypersensitive to UV-B, the more physiologically relevant range of wavelengths (Stapleton, 1992). In both screens, irradiated plants...
were subsequently kept in the dark (Britt et al.) or in gold light (Harlow et al.), both conditions that prevent CPD photoreactivation. Thus, both screening procedures would eliminate mutations that affect photoreactivation specifically. The two screening procedures have the potential to yield different types of mutants because meristem cells may rely more on recombinational repair mechanisms than does leaf tissue, which is relatively quiescent. It turns out, however, that both uvrI leaves and uvrH seedlings are hypersensitive to UV-B, so both mutations probably affect processes that occur in both dividing and quiescent cells.

Theoretically, a UV-hypersensitive mutant could arise in any number of ways—from a defect in protective pigment synthesis (or its induction by UV radiation) or DNA repair or from a mutation that renders a structural component of the cell more sensitive to UV than normal. Because both the uvrH and uvrI mutants sustain the same amount of pyrimidine dimer damage as their respective progenitors in response to a given dose of UV, neither mutation is likely to increase the transparency of the plant to UV radiation. In addition, both mutants are able to photoreactivate CPDs at normal rates, a result consistent with the fact that both were isolated under nonphotoreactivating conditions. Dark repair (i.e., excision plus postreplication repair) of CPDs also appeared normal in uvrI mutants.

Both mutants do, however, appear to be defective in DNA repair. By analyzing the repair of (6-4) photoproducts using antibodies specific for these lesions, Britt et al. (1993) found that whereas this type of pyrimidine dimer is repaired rapidly in plants of the progenitor strain, it is repaired slowly, if at all, in the uvrI mutant. Evidence that uvrH is defective in a different repair process comes from Harlow and coworkers’ observation that mutant plants were able to tolerate higher doses of UV when they were transferred to white (photoreactivating) light after irradiation than when they were transferred to gold light. Because photoreactivation could increase the UV resistance of the mutant, and assuming that CPD cleavage is the only repair process that is enhanced appreciably by photoreactivating light, the uvrH mutant is probably defective in dark repair of CPDs. By contrast, mutants defective in (6-4) photoproduct repair should show the same phenotype whatever the light conditions to which they are exposed after UV irradiation. The assumption that photoreactivation does not enhance (6-4) photoproduct repair in plants has yet to be verified, however, and it is possible that the uvrH mutation will turn out to affect this or some other repair process that is found to be light activated.

Harlow and coworkers’ further characterization of the uvrH mutant shows that it is hypersensitive not only to UV radiation but also to γ-irradiation. Ionizing radiation and chemicals such as bleomycin produce double-strand DNA breaks, which cannot be corrected by excision repair. In yeast, two main groups of mutants are hypersensitive to ionizing radiation. The RAD6 epistasis group, mutants of which are abnormally sensitive to a wide range of DNA-damaging agents, includes genes that appear to be involved in error-prone repair pathways, whereas genes of the RAD52 epistasis group are required for recombinational repair of DNA breaks caused by ionizing radiation (Friedberg, 1988). Thus, like mutants in these two epistasis groups, uvrH may be defective in a postreplication repair process. However, it is not clear how such a defect would affect mature leaf cells, which are largely nondividing. One possibility is that postreplication repair mechanisms are active even in nondividing cells, if only in chloroplasts. Alternatively, it is possible that UVH plays a role in both excision and postreplicative repair processes. Precedent for this comes from the observation that although genes of the yeast RAD3 epistasis group are involved primarily in excision repair, certain of them are also involved in recombinational repair (Friedberg, 1988).

Although genetic studies of plant DNA repair are just getting underway, it is clear that, together with biochemical studies, they will provide essential insights into the mechanisms by which plant cells detect UV radiation and cope with UV-induced damage. Only as we begin to find answers to such questions as: what sort of photoreceptor detects and measures UV? How is this detection transduced to activate flavonoid biosynthesis and, possibly, repair mechanisms? Do plants, which after all are exposed to light more than most animals, have unique repair mechanisms? What plant processes are particularly sensitive to UV radiation? Will we be able to predict how higher plants will be affected by potential increases in the amount of solar UV reaching the earth’s surface. The extent of ozone layer depletion may still be a matter of debate, but the effect of UV radiation on plants need not be.


A Ray of Light on DNA Repair

R. Chasan

Plant Cell 1994;6;159-161
DOI 10.1105/tpc.6.2.159

This information is current as of October 13, 2017