On the Inside

Suppression of a Soybean (Glycine max) Seed Allergen

Soybean allergy can affect humans as well as farm animals. Gly m Bd 30 K, a member of the papain protease superfamily, is a major soybean allergen. **Herman et al. (pp. 36–43)** used transgene-induced gene silencing to prevent the accumulation of Gly m Bd 30 K protein in soybean seeds. The Gly m Bd 30 K-silenced plants and their seeds lacked any compositional, developmental, structural, or ultrastructural phenotypic differences when compared with control plants. Proteomic analysis of extracts from transgenic seed detected the suppression of Gly m Bd 30 K-related peptides but no other significant changes in polypeptide pattern. Critics of genetically modified crops have raised the possibility that the genetic modification of plants by transgenic methods could potentially introduce novel protein allergens into foods. However, as Herman et al. demonstrate, it is also possible to remove a major food allergen by gene-silencing techniques. Thus, biotechnology offers the prospect of eliminating many allergens that pose difficulties for sensitive people.

Control of Tomato (Lycopersicon spp.) Fruit Size

A key morphological change that has accompanied the domestication of many fruit and vegetable crops including tomato has been the dramatic expansion of fruit and explosion of shape variation (Fig. 1). The wild forms of tomato bear small (approximately 1–2 g), round, seed dense berries—ideal for reproduction and dispersal. In contrast, cultivated tomatoes typically produce fruit that weigh anywhere from 50 to 1,000 g, come in a wide variety of shapes (e.g. round, oblate, pear-shaped, torpedo-shaped), and are not well adapted for seed dispersal in the wild. Genetic studies in-

Figure 1. Certain domesticated tomato fruits (left) are dramatically larger than their wild counterparts (right). Liu et al. (Cornell University, Ithaca, NY) provide evidence that much of this size difference is attributable to a heterochronically regulated, negative fruit growth regulator (*fw2.2*) that affects cell division patterns in the tomato fruit. (copyrighted by Kent Loeffler)

volving crosses of wild and cultivated tomatoes have shown that most of the variation in size and shape can be attributed to fewer than 30 quantitative trait loci (QTLs), with a smaller subset of these accounting for a disproportionate amount of variation. One of the major QTLs involved in tomato domestication, *fw2.2*, is a negative fruit growth regulator that accounts for approximately 30% of the variance in fruit weight. The heterochronic expression of this allele in different tomato species is a major determinant of the fruit mass variation between wild and domesticated tomato species. In this issue, **Liu et al. (pp. 292–299)** report on their construction of a gene dosage series that exhibits a 7-fold range in *fw2.2* transcript accumulation. These lines were characterized for associated changes in fruit development, fruit anatomy, cell proliferation, fertility, and other reproductive parameters. Their results provide strong evidence for both the negative regulator and transcriptional control hypotheses and reveal that *fw2.2* exerts its effects by influencing cell division patterns in the pericarp and inner placental tissues.

Laser Capture Microdissection of Plant Cells

Methods such as immunolocalization, in situ hybridization, and reporter gene visualization have permitted the cell-specific analysis of the expression of individual genes and of the accumulation of individual proteins. There is a pressing need, however, to develop techniques that provide such information on a genomic and proteomic scale. Laser capture microdissection (LCM) is a technique by which individual cells can be harvested from tissue sections while they are viewed under the microscope, by tacking selected cells to an adhesive film with a laser beam. LCM provides a rapid means of isolating pure cellular preparations directly from heterogeneous tissues, based on conventional histological identification. Specific markers can assist with the identification of the desired cells, before or after isolation, but they are not a requirement for LCM itself. Harvested cells can provide DNA, RNA, and protein for the profiling of genomic characteristics, gene expression, and protein spectra from individual cell types. Most studies using LCM have thus far used animal tissues as subjects, and the reported methods for the fixation, sectioning, visualization, and extraction of macromolecules in LCM experiments have been based on protocols optimized for animal cells. In this issue, **Kerk et al. (pp. 27–35)** report upon their progress in optimizing LCM for a variety of plant tissues and species, permitting the harvesting of cells from paraffin sections that maintain histological detail. They show that RNA can be extracted from LCM-harvested plant cells in amounts and qualities that are sufficient for the comparison of RNAs among individual cell types. The linear amplification of LCM-captured RNA should permit the expression profiling of plant cell types.

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Programmed Cell Death in Dunaliella

In higher plants, programmed cell death (PCD) mediates the hypersensitive response in plant-pathogen interactions, floral and organ abortion, senescence, aerenchyma formation, and the differentiation of tracheary elements. Surprisingly, cell cultures of the unicellular chlorophyte alga *Dunaliella tertiolecta* undergo PCD less than a week after being placed in darkness. Upon cell death, the cells literally dissolve, and the culture, which on the previous day had been green, becomes transparent. In this issue, **Segovia et al. (pp. 99–105)** report that the cell death program in *D. tertiolecta* is associated with caspase-like activity and that the morphology and biochemical features of the dying algal cells resemble PCD in metazoan cells. The cell death phenomenon in *D. tertiolecta* confers no obvious ecological or evolutionary fitness. This alga cannot use the dissolved organic compounds released from lysis for its own growth, and the organism does not reproduce sexually; hence, suggestions that cell death has evolved for "altruistic" functions or for cellular differentiation cannot be invoked in this organism. The authors support the idea that the PCD machinery of extant eukaryotes may not been "invented" for this function, but rather have been recruited from proteins that in unicellular organisms perform regulatory functions. Although the caspase-like proteins in *D. tertiolecta* may normally serve some housekeeping purposes, they also become maladaptively activated during dark-induced starvation. The authors hypothesize that key elements of cell death pathways may have been transferred to the nuclear genome of early eukaryotes through ancient viral infections in the Precambrian Ocean before the evolution of multicellular organisms and were subsequently appropriated into both metazoan and higher plant lineages.

Polychlorinated Biphenyl Bioremediation by Rhizosphere Microbes

Polychlorinated biphenyls (PCBs) are especially problematic organic pollutants because of their toxicity to a variety organisms, their rapid movement in the ecosystem, their high persistence, and their ability to accumulate in the food chain. Although PCB-degrading bacteria are found ubiquitously in the environment, most are inefficient in degrading PCBs. The major cause for this seems to be the lack of sustaining nutrients in the near-starvation conditions found in the soils, including the rhizosphere. Hence, the primary challenge for successful bioremediation of PCB-contaminated soil is to devise methods to encourage the growth (leading to more efficient PCB-removal) of a select species of microbes, which either are indigenous to PCB-contaminated sites or are introduced to these sites. Using a "rhizosphere metabolomics" approach, **Narasimhan et al. (pp. 146–153)** show that phenylpropanoids constitute 84% of the secondary metabolites exuded from Arabidopsis roots. They demonstrate enhanced depletion of PCBs by Arabidopsis root-associated microbe strains (*Pseudomonas* spp.) that have a nutritional advantage over other soil bacteria in their heightened ability to metabolize phenylpropanoids. One of these strains was able to remove more than 90% of the PCBs within the rhizosphere within 2 weeks. The strategy of enhancing the soil microbial populations using natural secondary metabolites exuded by plants could potentially be applied to the removal of many classes of pollutants in vegetated soils. In those cases where the pollutant-degrading microbes are not known to use secondary metabolites, such properties could potentially be introduced by genetic engineering.

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