Genetically Engineered Plants and Foods: A Scientist's Analysis of the Issues (Part II)

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Abstract

Genetic engineering provides a means to introduce genes into plants via mechanisms that are different in some respects from classical breeding. A number of commercialized, genetically engineered (GE) varieties, most notably canola, cotton, maize and soybean, were created using this technology, and at present the traits introduced are herbicide and/or pest tolerance. In 2007 these GE crops were planted in developed and developing countries on more than 280 million acres (113 million hectares) worldwide, representing nearly 10% of rainfed cropland. Although the United States leads the world in acres planted with GE crops, the majority of this planting is on large acreage farms. In developing countries, adopters are mostly small and resource-poor farmers. For farmers and many consumers worldwide, planting and eating GE crops and products made from them are acceptable and even welcomed; for others GE crops raise food and environmental safety questions, as well as economic and social issues. In Part I of this review, some general and food issues related to GE crops and foods were discussed. In Part II, issues related to certain environmental and socioeconomic aspects of GE crops and foods are addressed, with responses linked to the scientific literature.

Contents

1. INTRODUCTION

Genetically engineered (GE) crops and foods have been commercially available in the United States since 1995 and their adoption around the world followed, showing increases each year since their introduction (**Table 1**). Whereas the majority of the acreage is in the United States, most farmers who grow these crops reside outside the United States—more than 10 million of the 12 million adopters are in developing countries (165). These GE crops created by recombinant DNA (rDNA) have been overwhelmingly accepted by farmers, but some consumers remain skeptical. In Part I of this review (186a), general descriptions of the process of genetic engineering, its implications, and its regulation were discussed, as well as responses to several food and food safety issues. In Part II certain environmental and socioeconomic issues are discussed. Not all issues that have been raised are discussed and not all aspects of the issues reviewed are addressed, but the present state of knowledge is reviewed.

As scientifically accurate a picture as possible was presented by linking responses to peer-reviewed literature. This approach does not imply that people possessing the same scientific information will come to the same conclusions about GE crops and their products. Individual value judgments vary and thus different conclusions are reached. As a scientist, I feel, however, that what science has discovered about these crops should be a part of what individuals consider in making decisions about growing and consuming these crops.

2. ENVIRONMENTAL ISSUES

When contemplating environmental impacts of GE crops, it is important to consider that the fundamental issues raised are similar in many ways to those encountered with crops created by other genetic modification methods, such as mutation or marker assisted selection, and cultivated in other ways, such as the use of integrated pest management and organic and biocontrol methods. The issues include the nature of the genetic change, the impact on the genotype and phenotype of the crop, and the consequences for the environment. Regarding these issues, it was recently suggested that certain analyses are too crude to allow meaningful assessments of environmental consequences and require geographical data to be collected on a smaller, more defined basis (197).

2.1. Will the Widespread Use of *Bt* Crops Lead to the Development of Insect Resistance to *Bt*?

Bacillus thuringiensis (Bt), a widespread soil bacterium, produces insecticidal proteins called Bt toxins (127). There are many Bt strains that produce characteristic sets of toxins, each with its own activity spectrum that targets larvae of specific insect species. For example, some Bt toxins kill larvae of particular species of moths and butterflies; others kill larvae of certain species of beetles or mosquitoes. Bt sprays have been used to control insects since the 1920s (127), but use of specific Bt toxins has increased dramatically since 1996 with the introduction of GE crops.

Bt toxins are also called Cry toxins because they exist as crystals inside the bacterium. Full-length Cry toxins are inactive until cleaved to generate their active form in the insect midgut (236, 261). Binding of activated forms of Cry toxins to receptors in the midgut is generally believed to be essential for toxicity. According to one model (168, 277), after binding to midgut receptors, activated toxins form oligomers that create pores in midgut membranes, causing contents to leak, ultimately killing the larvae. The precision of Bt proteins for certain insects and their lack of effects in mammals are due to the specificity of receptor binding (107).

As of July 2008, deregulation has been approved in the United States for thirteen different Bt events of corn (*Zea mays*), five of cotton (*Gossypium hirsutum*), five of potato (*Solanum tuberosum*), and one of tomato (*Lycopersicum esculentum*) that produce one or more Cry proteins: Cry1Ab, Cry1Ac, Cry1F, Cry2Ab, Cry3A, Cry3Bb1, Cry9C, Cry34Ab1,

GE: genetically engineered

Recombinant DNA (rDNA): DNA that is manipulated enzymatically in the laboratory using recombinant DNA technologies

Peer-reviewed: A publication that has been reviewed, usually anonymously, and approved by other experts in the author's field before it is published

Marker assisted selection: process by which breeders use a marker (morphological, biochemical, DNA, or RNA variation) for indirect selection of a genetic determinant for a trait of interest

Phenotype: observable physical or biochemical characteristic of an organism dictated by genetic makeup or environmental effects

Bt: *Bacillus thuringiensis*

Cry: crystal protein

Toxicity: adverse physiological effects following exposure to a substance

Hectare: a metric unit used for area measurement (especially in agriculture) that is equivalent to approximately 2.471 acres or 107,639 square feet

APHIS: Animal and Plant Health Inspection Service

USDA: United States Department of Agriculture

EPA: Environmental Protection Agency

Pyramid (stacking) strategy: deployment of varieties expressing different genes by incorporating both genes in a single variety, as with two different *Bt* genes

and Cry35Ab1 (159). Bt potato and tomato are not grown commercially in the United States at present. Bt corn producing Cry1Ab and Bt cotton producing Cry1Ac account for the majority of the 494 million acres (200 million hectares) (hectare = ha = 2.47 acres) of Bt crops grown worldwide during the ten-year period from 1996 to 2007 (164). These two Bt crops kill some key lepidopteran pests, including European corn borer (*Ostrinia nubilalis*) on maize and pink and cotton bollworm (*Pectinophora gossypiella* and *Helicoverpa armigera*) and tobacco budworm (*Heliothis virescens*) on cotton. As of July 2008, applications have been filed with the Animal and Plant Health Inspection Service (APHIS) of the U.S. Department of Agriculture (USDA) to conduct 844 small-scale, precommercial field tests of 30 different plant species engineered with Bt genes [e.g., apple, cranberry, grape, peanut, poplar, rice, soybean, sunflower, and walnut (158)], although the actual number of tests conducted is not known.

Evolution of insect resistance to Bt toxins can reduce the long-term effectiveness of Bt crops (136, 282, 286, 290). Strains of many pests have been selected for resistance to Bt toxins in the laboratory, and two lepidopteran insects, *Plutella xylostella* and *Trichoplusia ni*, have evolved resistance to Bt sprays in the field and in greenhouses, respectively (166, 284). The primary strategy in the field for delaying insect resistance to Bt crops is planting refuges of non-Bt crops near Bt crops (103, 136, 286). This strategy is based on the idea that insects feeding on plants in the refuge are not selected for resistance, because those plants do not make Bt toxins. Under ideal conditions, insect resistance to Bt toxins is recessive. Thus, heterozygous offspring, produced when homozgygous resistant insects mate with susceptible insects, are killed by the Bt crop. Models predict that resistance can be postponed substantially if the rare homozygous resistant insects surviving on a Bt crop mate with the more abundant susceptible insects from refuges (136, 282). The strategy is called the high-dose/refuge strategy because the created plants produce Bt toxin concentrations high enough to kill heterozygous insects, thereby making resistance functionally recessive (286).

In the United States and some other countries, refuges of non-Bt crops are required (103). A 2005 survey showed that U.S. farmers believe refuges are effective in managing resistance (6); 91% of farmers were found to meet the regulatory requirements for refuges associated with Bt corn (218). A study of Bt cotton revealed compliance with the refuge strategy was higher than 88% in five of six years from 1998 to 2003 (63). In addition to mandating non-Bt crop refuges, the U.S. Environmental Protection Agency (EPA) requires monitoring for field resistance to provide early warning of resistance development (103). In Arizona, where Bt cotton producing Cry1Ac has been used widely since 1997 and pink bollworm has been under intense selection for resistance, a statewide surveillance system for resistance exists. From 1997 to 2004, results of laboratory bioassays of insects derived annually from 10 to 17 cotton fields statewide showed no net increase in mean frequency of pink bollworm resistance to Bt toxin (284). DNA screening from 2001 to 2005 also showed that resistance-linked mutations remained rare in pink bollworm field populations (285). Sustained efficacy of Bt cotton has contributed to long-term regional suppression of pink bollworm (64).

Although the strategies implemented to delay resistance have helped sustain efficacy of Bt crops longer than many scientists expected, field-evolved resistance to Bt crops was reported recently (200, 286, 312). Analysis of published monitoring data from the United States, Australia, China, and Spain for major lepidopteran pests targeted by Bt crops indicated field-evolved resistance in *Helicoverpa zea*, but not in pink bollworm or the four other insects examined (*Helicoverpa armigera*, *Heliothis virescens*, *Ostrinia nubilalis*, and *Sesamia nonagrioides*). Evaluation of the large data sets of two landmark studies (7, 192) revealed that resistance to Cry1Ac produced by Bt cotton occurred in 2003 to 2004 in some field populations of *H. zea* in Arkansas and Mississippi, but not in *H. virescens* from the same region. Resistance of

H. zea to Cry1Ac has not resulted in widespread crop failures, in part because existing insecticide sprays and other tactics are still effective against this pest (286). Correspondence between monitoring data and results from computer modeling of resistance evolution suggests that the principles of the refuge strategy for these pests and Bt crops are relevant in the field. Also consistent with monitoring data, modeling suggests *H. zea* would evolve resistance faster than other pests, because its resistance to Cry1Ac is dominant, not recessive as with other Bt toxins (286). Monitoring data also suggest relatively large refuges may have delayed *H. zea* resistance to Cry1Ac in North Carolina (286). Field resistance of *Busseola fusca* was reported in 2007 to Cry1Ab and Cry1F in maize in South Africa (312), and in 2008 field resistance of *Spodoptera frugiperda* was reported in Puerto Rico (200).

First-generation GE crops produced only one Bt toxin in each plant. A second approach designed to delay resistance is called the pyramid or stacking strategy and entails combining two or more toxins in a single plant, each with different modes of action (254). If no cross-resistance exists between the two toxins, frequency of insect resistance to both toxins is much lower than that for one toxin. Importantly, tests of this approach with a model system using GE broccoli and the insect pest *Plutella xylostella* suggested that concurrent use of plants with one and two toxins selects for resistance to two-toxin plants more rapidly than the use of two-toxin plants alone (338). In the United States, Bollgard II® cotton producing Cry1Ac and Cry2Ab was introduced commercially in 2003 and has been grown alongside Bollgard cotton producing only Cry1Ac. On the basis of the results with the model broccoli system (337), this concurrent use of one-toxin and two-toxin Bt cotton may not optimize the benefits of the two-toxin cotton. In contrast, Australian cotton growers stopped planting cotton that produces only Cry1Ac soon after two-toxin Bt cotton became available; this strategy might result in delayed resistance development (337).

Other approaches to delaying resistance development have been suggested. The efficacy of mixing seeds of Bt and non-Bt varieties of the same crop has been debated (193); to date, evidence to resolve this issue has been limited to theoretical models and small-scale experiments (148, 187, 193, 267, 283). The practical advantage of seed mixtures in ensuring that non-Bt plants grow near Bt plants may outweigh possible advantages of spatially separate refuges (65). Another suggestion to shorten periods of insect exposure and slow evolution of insect resistance is the use of inducible promoters to drive Bt gene expression only during insect attack (31). Another approach uses knowledge of insect resistance mechanisms to design modified toxins to kill resistant insects (277), on the basis of the fact that the most common mechanism of Cry1A resistance in lepidopteran insects involves disruption of Bt toxin binding to midgut receptors (111). Mutations in midgut cadherins that bind Cry1Ac are linked with and probably cause resistance to Cry1A toxins in at least three lepidopteran pests of cotton (121, 209, 333). The role of cadherin in Bt toxicity was elucidated by silencing the cadherin gene in *Manduca sexta*, which reduced its susceptibility to Cry1Ab (277). Consistent with the role of cadherin in promoting toxin oligomerization demonstrated by removing an α-helix from Cry1A toxins, toxin-binding fragments of cadherin were required for oligomer formation of native Cry1A toxins, but not for Cry1A toxins lacking the α-helix. The modified Cry1A toxins killed cadherin-silenced *M. sexta* and Cry1A-resistant pink bollworm larvae, suggesting that modified Bt toxins might be effective against insects resistant to native Bt toxins (277).

In summary, just as insects have evolved resistance to synthetic insecticides and Bt toxins in sprays, they are evolving resistance to Bt toxins in GE crops. The elapsed time before the first cases of field resistance of insects to Bt crops were reported has been longer than what was predicted under worst-case scenarios, suggesting that management strategies may have delayed resistance development. Despite

RNAi: RNA interference

Pesticide: any naturally occurring or synthetically produced substance or mixture of substances used to prevent, destroy, repel, or mitigate any pest, which includes insects, weeds, fungi, bacteria, viruses, or mice and other animals

documented cases of resistance, Bt crops remain useful against most target pests in most regions. As insect resistance to Cry toxins currently deployed in Bt crops increases, other strategies to create GE crops resistant to insects are being developed, including vegetative insecticidal proteins (Vips) from Bt (190) and RNA interference (RNAi) (32, 195).

2.2. Can Genetically Engineered Crops Cause Adverse Effects on Nontarget Organisms?

Various published studies analyzed effects of Bt maize on nontarget insects. Two well-known studies focused on monarch butterflies (191) and on black swallowtails (329). The first, a note to *Nature* in 1999, was a laboratory study in which monarch caterpillars were fed milkweed leaves dusted with loosely quantified amounts of pollen from a single Bt corn variety. In the second study in 2000, black swallowtail caterpillars were placed different distances from a cornfield planted with a Bt corn variety different from that used in the 1999 study; populations were studied for effects of Bt for seven days. In the first study more monarch caterpillars died when they ate leaves dusted with Bt corn pollen versus leaves dusted with conventional corn pollen. In the second study, no negative effects of Bt pollen were found on numbers of swallowtail caterpillars.

After those papers appeared, data from numerous university studies performed in the laboratory and in the field on the effects of Bt corn on monarch butterflies were published (62; for a summary of studies see References 102 and 264). After reviewing the data, the U.S. EPA concluded there was a very low probability of risk to monarch butterflies beyond 12 feet from the Bt corn field. Two varieties, Bt11 and Mon810, had no acute adverse effects, even at pollen densities greater than the highest densities observed in cornfields (239). Another variety, 176, had limited negative impacts on some nontarget insects because expression of the 3′ truncated *cry1Ab* was linked to a maize pollen-specific promoter (2). Rates of larval survival and weight gain in fields of 176, however, were much greater than in fields sprayed with the insecticide Warrior 1E (279). The EPA concluded from these studies that Bt corn was not a significant factor in field death of monarch larvae, particularly relative to factors such as the widespread use of pesticides and destruction of the butterfly's winter habitats (207, 237).

To "encourage evidence-based risk analysis," Marvier et al. (198) published a report in 2007 describing a searchable database on the effects of Bt on nontarget insects (217). In a meta-analysis of 42 field experiments, taking into account location, duration, plot sizes, and sample sizes, these authors concluded that (*a*) the mean abundance of all nontarget invertebrate groups, in terms of numbers, survival, and growth, was greater in Bt cotton and Bt maize fields than in non-Bt fields managed with insecticides but, (*b*) if Bt crop fields and insecticide-free fields were compared, certain nontarget insects were less abundant in Bt fields.

Effects of Bt on the biodiversity of nontarget soil microorganisms were studied following four years of cultivation of four maize varieties with two different Bt proteins (Cry1Ab and Cry3Bb1) versus near isogenic non-Bt varieties (154). In general, although numbers and types of microbes and enzyme activities differed from season to season and among varieties, no statistically significant differences were seen in numbers of different microbes, enzyme activities, or pH between soils with Bt and non-Bt corn. In similar studies comparing impacts on the rhizosphere of Bt cotton versus non-Bt cotton, various enzymatic activities were measured before and after harvest (268). The authors concluded that richness of the microbial communities in the rhizosphere did not differ between Bt and non-Bt cotton. No Cry2Ab protein was detected in the rhizosphere soil of field-grown Bt rice (316).

Effects on foliage-dwelling arthropods of Bt maize expressing Cry3Bb1 to protect against corn rootworm (*Diabrotica* sp.) were compared with those of conventional insecticide treatments (43). Bt maize had no consistent adverse impacts on abundance of any nontarget arthropods; however, insecticide treatments applied to the plant foliage significantly and consistently decreased abundance of three nontarget insects: ladybird beetles, lacewings, and damsel bugs. Thus, reducing foliar sprays with the use of Bt corn has the potential to enhance approaches using biological control agents.

Another potential effect of Bt crops on nontarget organisms is the passage of Bt from fields to nearby aquatic environments with the possibility of increasing horizontal gene flow to microbes and mortality of nontarget stream insects. To test this potential effect, soil, sediment, and water samples were analyzed after spiking sediments and surface waters with *Bacillus thuringiensis kurstaki* and genomic DNA from GE Bt corn (89). PCR analyses revealed that half-lives for both sources of Bt DNA were 1.7 d for clay- and sand-rich sediments and 14.3 d in surface water. Soil, sediment, and surface water from Bt maize fields were also tested for the presence of *cry1Ab* two weeks after pollen release, after corn harvest, and after mechanical root remixing. Sediments had more *cry1Ab* DNA than surface water, perhaps reflecting binding to soil particles that increased its persistence; however, Cry1Ab protein was undetectable in most samples. Without making field measurements on nontarget populations, it was suggested that release of products with Bt transgenes into the environment might adversely affect nontarget organisms; however, other researchers objected because actual measurements were not made (35, 232).

Although many studies focus on potential negative effects of Bt on nontarget organisms, potential benefits to nontarget insects have also been noted. Bt maize is more susceptible to corn leaf aphids (*Rhopalosiphum maidis*), which leads to larger colony densities and increased production of the honeydew consumed by beneficials such as a parasitoid of aphids, *Cotesia marginiventris* (106). This observation underscores the delicate balance in nature between beneficial and detrimental side effects of insect protection strategies.

2.3. Could the Use of Genetically Engineered Crops Result in a Loss of Plant Biodiversity?

Food crops were first domesticated from wild species approximately 10,000 years ago when nomadic hunter-gatherers shifted to an agrarian lifestyle (287). Through human involvement in plant selection a profound effect was exerted on the genetic landscape, as plant species with favorable mutations were selected for propagation. Biodiversity in agroecosystems, which reflects not only species richness, but also the diversity of their interactions (214), continued to decline with changes in agricultural practices and plant breeding efforts, both of which focused on providing the high yields demanded by expanding populations (8, 101, 274). These negative effects on biodiversity, sometimes termed genetic erosion (116), also led to loss of weed species, killing of nontarget pests, and destruction of natural habitats for insects and wild animals (204). The larger the agricultural acreage, the greater the impact on surrounding flora and fauna.

Frankel (116) established principles of genetic erosion that describe agriculture's impact on biodiversity: (*a*) during premodern agriculture, in centers of diversity, crop species were stable; (*b*) introduction of modern agricultural technologies, including new varieties, led to instability; (*c*) competition between local and introduced varieties led to displacement of local varieties; and (*d*) displacing local varieties eroded genetic variability of regional crop populations. Plant breeding in the early 1960s produced high-yield varieties of major food crops, resulting in yield increases but also significant displacement of traditional varieties and a concomitant loss in genetic diversity, particularly landraces of cereals and legumes (100).

Recognition of this consequence on genetic diversity led to the development of global genebanks and collections to conserve genetic resources, such as those maintained by the USDA's National Plant Germplasm System and the Consultative Group on International Agricultural Research (CGIAR). These

Transgene: gene that is manipulated using recombinant DNA technologies and reintroduced into a host organism, where the DNA becomes part of the host's genetic makeup and is passed to the next generation

Genetic erosion: loss of genetic diversity between and within populations of the same species over time or reduction in the genetic bases of a species due to human intervention, environmental changes, and other factors

Germplasm: A collection of genetic resources of an organism, sometimes stored as a seed collection

CGIAR: Consultative Group on International Agricultural Research

Classical breeding: deliberate crossing of compatible individuals to introduce traits/genes from one organism into a new genetic background

Gene flow: transfer of genetic information between individuals or populations; can occur in plants when pollen moves from one compatible plant to another

HT: herbicide tolerant

IP: intellectual property

DEFRA: Department for Environment Food and Rural Affairs

collections, which preserve precious landraces and wild relatives, are the foundation of future classical breeding, marker assisted selection, and genetic engineering efforts and it is critical to maintain and enlarge these resources. Molecular and genomic technologies enable identification of genetic variants and development of detailed genetic descriptions of diversity, leading to greater appreciation of these resources. Information technology, which enables analysis of large data sets, has also led to advances in conservation and use of plant genetic resources.

The commercialization of herbicide-tolerant (HT) and insect-tolerant (Bt) GE crops raised questions about the environmental and genetic conservation impacts of gene flow from GE crops to wild and weedy relatives. This gene flow could lead to selective advantages (e.g., enhanced invasiveness and/or weediness) of recipients in certain environments (145); this phenomenon is of particular interest in centers of crop diversity. Careful measures are needed when cultivating GE crops near such centers (100); however, this situation is not unique to GE plants and can and does happen with conventionally bred, commercialized crops (97). Key to judging the impact of transgene movement is the nature of the trait and the frequency of its introduction into an ecosystem. Studies of the impact of transgenes moving into wild relatives and the potential to change ecosystem dynamics are currently requested in environmental impact statements written for commercial release of a new GE plant (10). Although such tests are limited in scope and do not address all eventualities, they do provide insights into possible outcomes. Just as with other agricultural practices, certain impacts of GE crops on the environment need to be monitored, even after deregulation (Section 2.1).

Another consideration regarding effects of GE crops on diversity of local, adapted crop varieties is that most current, commercial GE varieties were developed by large, mostly international companies. The few small seed companies remaining (110) have no legal access to GE traits, due in part to intellectual property (IP) issues (Section 3.3). Thus it is difficult for these companies to move traits into local varieties, which leaves the task to larger companies. However, regulatory costs, IP, and other issues likely limit the numbers of varieties into which GE traits are moved by these companies, potentially narrowing the genetic base available to farmers.

2.4. Will the Use of Genetically Engineered Crops Result in the Population Decline of Other Organisms?

The diversity and numbers of other organisms known to play important roles in controlling pests and diseases (e.g., microorganisms, predators, birds, parasitic wasps) could be affected by the presence of GE crops. On the basis of mathematical modeling predictions, the diversity of such organisms might be at risk because of HT crops (319). Assuming fewer weeds grow in HT crop fields versus conventionally sprayed fields, smaller numbers of weeds might lead to less food for grain-eating birds and to possible declines in bird populations, a potential problem with large-scale deployment of HT crops. Conversely, because herbicides are usually applied later with HT crops, there are possible advantages for birds that breed in such fields (99), because dead weed material left behind can serve as nesting grounds (112). This potential consequence of HT crops may not be as significant in North America, where vast areas of unfarmed land remain for birds and animals to find weeds and seeds. In Europe, however, where land is more restrictive, the impact of HT crops might be more pronounced, leading to use of buffer strips to enhance feed and nesting habitats.

To quantify effects of HT crops in the United Kingdom on bird and animal populations, the impact of four HT crops (sugar beet, maize, and spring and fall oilseed rape) on abundance and diversity of farmland wildlife was compared with that of conventional varieties. This effort was initiated in 1999 when the Department for Environment, Food, and

Rural Affairs (DEFRA) commissioned an independent consortium of researchers to conduct a five-year study involving 266 field trials (87). Results of the study were not uniform for all crops. For sugar beet and spring rape, conventional varieties harbored more insects because of the presence of weeds and weed seeds. Growing HT maize led to more weeds and seeds because of late timing of herbicide application and thus resulted in more butterflies and bees. HT and conventional winter rape were comparable in numbers of weeds, but in HT rape there were fewer beneficial weeds, resulting in fewer bees and butterflies. Negative effects of HT weed control strategies on sugar beet can be counteracted by leaving two rows per 100 untouched, resulting in weed seed production equal to that in non-HT crops (234). Researchers concluded that differences among crops were not caused by plants' being GE, but instead were the result of HT varieties giving farmers new weed control options with differing impacts. In thinking about options, farmers should consider sizes of acreages planted, proximity to other crops, and crop rotations.

2.5. Will the Use of Herbicide-Tolerant Genetically Engineered Crops Lead to Superweeds?

The concept of a superweed conjures up the image of a weed taking over entire ecosystems, undeterred by existing herbicides. Although this scenario is not based in fact, problems with herbicide-resistant weeds are real, but not new. These problems have occurred with traditionally bred crops, as well as with HT GE plants. Historically herbicide resistance arose because of herbicide overuse or movement of conventional herbicide-tolerance traits to weedy species, resulting in plants not controllable with previously applied herbicides (58, 163, 281). Although this phenomenon does not lead to so-called "environmental disasters," it reduces the effectiveness of certain weed control strategies and decreases weed management options. Good weed management practices can ameliorate this situation in conventional, organic, and GE cropping systems (122, 141).

Resistant weeds have arisen associated with an herbicide used with a GE HT crop, i.e., glyphosate or Roundup® (216). Among the species worldwide with documented resistance are ryegrass (*Lolium perenne*) in Australia (240), goosegrass (*Eleusine indica*) in Malaysia (185), liverseed grass (*Urochloa panicoides*) in New South Wales (242), and in the United States horseweed (*Conyza canadensis*) in Delaware (311), California (272), Indiana, and Ohio (243) and pigweed (*Amaranthus palmeri*) in Georgia (82). Overuse of single herbicides can lead to this situation and will reduce the effectiveness of the GE HT crop. Having HT cultivars with resistance genes for herbicides with alternate modes of action that can be used in rotation will slow resistance development in weeds. Use of Liberty Link® varieties, which are tolerant to glufosinate (33), and development of GE dicamba resistance strategies (38) are steps in that direction (Section 2.6).

HT weeds can also arise because of outcrossing with HT GE crops. The frequency of occurrence depends on many factors, particularly the existence of compatible weedy species. In the United States some commercialized GE crops do not have native wild weedy relatives (142), but some do. Canola, in particular, can naturally form crop-wild hybrids and, even though fertility is often reduced, fertile offspring can be recovered (203, 318). For example, in Quebec, Canada, hybridization between transgenic canola (*Brassica napus*) and neighboring weedy *Brassica rapa* was documented (318). Although hybrid lineages declined dramatically over time, *B. napus* amplified fragment length polymorphism (AFLP) markers persisted in *B. rapa*, likely because of the presence of the HT transgene in diploid *B. rapa*, which, despite reduced pollen fertility, still produced offspring with high pollen fertility. More notably, the HT transgene persisted in the *B. rapa* population without herbicide applications from 2003 to 2008 (317). Also, a triple-resistant canola plant (146) with two GE traits and one mutation-induced HT

Outcrossing: process by which plants reproduce by dispersing their pollen to other compatible plants, rather than self-pollinating

Hybridization: crossbreeding plants of different varieties, species, or genera to create a plant with traits from each parent

AFLP: amplified fragment length polymorphism

GURT: genetic use restriction technology

EPSPS: 5-enol-pyruvylshikimate-3-phosphate synthase

PAT: Phosphinothricin-N-acetyltransferase

trait was found in Canada in 2000. HT *B. rapa* and the multiply resistant canola are controllable with other herbicides, but weed control options are reduced. Also, with triple-resistant canola, the likelihood for mixing GE canola with non-GE canola targeted for a GE-sensitive market is increased, possibly resulting in economic losses (Section 3.4). One approach to reducing transgene movement is use of genetic use restriction technologies (GURTs) (Section 3.2), which prevents gene passage to the next generation.

Overuse of herbicides can also result in weed shift, where weeds naturally resistant to an herbicide encroach upon areas where the herbicide is in use. In 2006, no Roundup®-resistant weed shifts had occurred with HT maize, but some had occurred with HT cotton (*Gossypium hirsutum*) and soybean (*Glycine max*), in some cases leading to economic concerns (81). Approaches to mitigate this situation include use of other herbicides with Roundup®, rotation to non-Roundup® herbicides, and/or use of non-Roundup Ready® crops. Development of either herbicide-resistant weeds or weed shifts with HT crops might negate the positive environmental benefits of HT crops. Weeds can also escape herbicide treatment on the basis of application rate, weed age and size, spray volume, adjuvants used, water quality, and interactions with other herbicides that affect efficacy (263). Weed escapes also occur because some weeds germinate late, after the last herbicide application (147), and thus are not controlled by Roundup®.

2.6. What Methods Are Used to Help Plants Protect Themselves Against Pests?

Pesticides, used to control plant pests, are needed because plants cannot move to avoid pests. Although useful in some cases, pesticides (i.e., herbicides, insecticides, and fungicides) are costly to the farmer and can be damaging to the environment and to humans. Herbicides must distinguish between desirable crop plants and undesirable weedy species, for example between commercial rice and red rice (both *Oryza sativa*); the latter is the most troublesome weedy species in many rice-growing regions of the world (323).

Crop tolerance to herbicides is achieved (*a*) by mutations that render a plant not susceptible to the herbicide or (*b*) through the introduction of transgenes. An example of the first approach was the identification of varieties that, after treatment with a chemical mutagen, were tolerant to imidazolinone herbicides; these Clearfield® varieties (80) are tolerant to herbicides, such as Pursuit® and Raptor®. In the second approach to be commercialized, GE crops, tolerant to glyphosate or Roundup®, were engineered with a bacterial gene encoding a target enzyme, 5-enol-pyruvylshikimate-3-phosphate synthase (EPSPS), which confers tolerance to the herbicide (30). More recently, other commercialized HT GE crops were created with tolerance to glufosinate or Liberty® by introducing phosphinothricin-N-acetyltransferase (*pat*) or bialaphos resistance (*bar*) genes from *Streptomyces* sp. that encode enzymes that detoxify the herbicide's active ingredient (324). The leading commercialized insect-tolerant GE crops have genes from the soil bacterium *Bacillus thuringiensis*, which encode pesticidal Cry proteins that protect the plant against specific insect pests (Section 2.1).

The first GE HT crops, cotton, corn, and soybean, have been grown commercially in the United States since 1995 (3). In 1996 HT soybean comprised 7% of total U.S. soybean acreage, compared with 92% in 2008 (**Table 1**; 299a). HT soybeans and cotton are the most widely and rapidly adopted GE crops in the United States, followed by insect-resistant Bt cotton and corn, which were also approved for commercial production in 1995. In 1996, Bt cotton was estimated to compose 15% of U.S. cotton acreage or 1.8 million acres (0.73 million hectares) (299), and Bt corn was grown on approximately 1% of the U.S. corn acreage (**Table 1**; 61, 109). Since 1996, both Bt corn and cotton crops have been widely adopted, and, as individual traits, represent 17% (298) and 18% (299), respectively, of cultivated U.S. acreage in 2008. However, these percentages represent varieties with individual traits and account for only part of the adoption because of stacked traits, i.e., introducing HT and Bt traits in the same plant (Section 2.1). In 2008 stacked varieties of corn made up 40% of acreage and stacked varieties of cotton comprised 45% of acreage; in combination with individual traits this adoption accounts for 80% of corn and 86% of cotton (**Table 1**). No stacked traits presently exist in commercial soybean varieties.

Table 1 Percent of total acreage of genetically engineered crops in the United States[1]

	Genetically engineered upland cotton			
	HT only	Bt only	Stacked	All
1996	2	15	NA[2]	**NA[2]**
1997	10	15	NA	**NA**
1998	26	17	NA	**NA**
1999	42	32	NA	**NA**
2000	26	15	20	**61**
2001	32	13	24	**69**
2002	36	13	22	**71**
2003	32	14	27	**73**
2004	30	16	30	**76**
2005	27	18	34	**79**
2006	26	18	39	**83**
2007	28	17	42	**87**
2008	23	18	45	**86**

	Genetically engineered maize			
	HT only	Bt only	Stacked	All
1996	3	1	NA[2]	**NA[2]**
1997	4	8	NA	**NA**
1998	9	19	NA	**NA**
1999	8	26	NA	**NA**
2000	6	18	1	**25**
2001	7	18	1	**26**
2002	9	22	2	**34**
2003	11	25	4	**40**
2004	14	27	6	**47**
2005	17	26	9	**52**
2006	21	25	15	**61**
2007	24	21	28	**73**
2008	23	17	40	**80**

	Genetically engineered soybean		
	HT only	Stacked	All
1996	7	NA[2]	**NA[2]**
1997	17	NA	**NA**
1998	44	NA	**NA**
1999	56	NA	**NA**
2000	54	0	**54**
2001	68	0	**68**
2002	75	0	**75**
2003	81	0	**81**
2004	85	0	**85**
2005	87	0	**87**
2006	89	0	**89**
2007	91	0	**91**
2008	92	0	**92**

[1] Data for 1996 to 1999 from Reference 109; data for 2000 to 2008 from USDA Economic Research Service for cotton (299), for maize (298), for soybean (299a). HT, herbicide tolerant; Bt, *Bacillus thuringiensis*.

[2] Data for stacked traits and total all genetically engineered varieties not available.

2.7. Does the Use of Genetically Engineered Crops Result in Decreased Use of Pesticides?

Having crops tolerant to herbicides and pest attack increases pest management options and can also reduce the number and strength of pesticide applications. Growth of GE HT crops also allows topical application of herbicides to crops and weeds, which replaces spraying between crop rows and mechanical removal of weeds, both of which can damage crops and result in environmental damage. Reducing mechanical tillage lowers fuel consumption and helps conserve soils prone to erosion and compaction (173). HT crops can also lead to more flexible herbicide treatment regimes.

Herbicide usage on HT GE crops has been analyzed in numerous studies. The National Center for Food and Agricultural Policy (NCFAP) published surveys in 2000, 2003, and 2004 on U.S. pesticide usage on GE crops by collecting information from industry experts, academic researchers, and Cooperative Extension. In 2004, HT canola, cotton, maize, and soybean, as well as Bt cotton and maize, were studied; reductions in herbicide active ingredient (AI) were 25 to 33% (259). In a 2006 publication, the USDA National Agricultural Statistics Service (NASS) looked at both herbicide and insecticide use, analyzing data up to 2002. AI use rates for HT cotton and corn and Bt corn declined from 1996 to 2002 (figure 8 in Reference 108); overall reductions in pesticide (herbicide + insecticide) use were observed as adoption of Bt and HT cotton, corn, and

AI: active ingredient

NCFAP: National Center for Food and Agricultural Policy

NASS: National Agricultural Statistics Service

Environmental impact quotient (EIQ): a relative value that estimates the environmental impact of a pesticide taking into account toxicity to wildlife, natural pests and humans, degree of exposure, aquatic and terrestrial effects, soil chemistry, etc.

Acre: a unit of surface area defined as an area 22 yards by 220 yards, equivalent to 4840 square yards or 43,560 square feet

EI: environmental impact

soybeans increased. This phenomenon led to an overall reduction of ca. 2.5 million pounds of AI, although slight increases in herbicide use with soybeans were found (109). The latter increase is consistent with the fact that, as glyphosate application to HT soybean acreage increased, concurrent shifts occurred toward less environmentally persistent herbicides (figure 2 in Reference 174), such as pendimethalin, trifluralin, and metolachlor (61). Taken together, these results agree with many field tests and farm surveys showing lower pesticide use for GE versus conventional crops (table 3 in Reference 108).

Using the same data from USDA NASS and other experts and extrapolating from trends when data were missing, another study also found higher glyphosate use from 2002 to 2004 on HT soybean compared with its use on conventional soybean but no increase from 1996 to 2001 (40). The increase in use from 2002 to 2004 was due in part to a switch to more effective herbicide mixtures and to more restrictive policies on herbicide use (174). Similar conclusions were drawn for HT maize. For Bt cotton, a trend was noted toward lower insecticide rates on conventional cotton, due in part to single Bt varieties needing bollworm-directed sprays late in the season. Lower pesticide use rates were observed for Bt maize; however, only part of conventional U.S. maize is normally treated with insecticides. The rate decreased in successive years, likely because of use of lower-rate insecticides such as cyfluthrin (40). A more promising approach for pesticide reduction for corn was the introduction in 2003 of a GE variety expressing a modified Cry3Bb1 protein (314), which protects against the western corn rootworm (*Diabrotica virgifera virgifera*), a difficult-to-control soil pest with a serious economic impact (206). To control rootworm, pesticides are often applied even when its presence is not known, because the economic impact of the pest is often not known until treatment is no longer effective; losses from rootworm damage are often high. Environmental implications regarding adoption of this variety should be considered; these include ecological effects on surrounding soils and the potential for rootworm to develop resistance (88).

The reason all reports on pesticide usage do not reach the same conclusions relates to the use of different data sets and/or different ways of calculating pesticide use (178). Disagreement exists on which methods are most accurate to calculate use rates, because they each reveal different aspects of herbicide usage. Regardless, certain parameters, such as agricultural practices used on and environmental conditions of the acreages compared, should be similar when comparing use rates.

Measuring amounts of pesticide AI used is helpful, but it does not provide adequate data on environmental effects (173), because each pesticide has different environmental and toxicological impacts. One means to take these factors into account uses the concept of an Environmental Impact Quotient (EIQ) (179). EIQ measures environmental and toxicological effects on the basis of many variables: toxicity of the AI, its mode of action, period of time AI persists, and ability of herbicide to contaminate groundwater. Each AI in a pesticide has a specific EIQ based on these parameters.

An EIQ Field Use Rating is determined by multiplying the EIQ value by (*a*) the amount of AI in a given amount of herbicidal product and (*b*) the amount of herbicidal product applied per acre. The smaller the EIQ Field Use Rating number, the smaller its environmental impact. By calculating EIQ Field Use Rates for each pesticide, impacts of different pesticides can be compared. EIQs can also be calculated for farm worker health, consumer health, and ecology (174).

In 2006 the environmental impact (EI) of cotton varieties expressing Cry1Ac and Cry2Ab was determined (177). Measurements of Bt protein expression, plant biomass, insecticide application rates, AI measurements, and insecticide EIQ values were used to produce an EI value, expressed as kilograms (kg) AI per hectare for conventional, single-gene, and two-gene Bt cotton from 2002 to 2003 and from 2003 to 2004. The average insecticide EI for conventional cotton was 135 kg AI/ha; for the two-gene

Bt variety this value was 28 kg AI/ha as a result of changes in both approach to insecticidal applications and reduction in usage. From 1997 to 2004 in Australia the EIQ method was used to study the EI of Bt cotton (the single-trait variety Cry1Ac and the double-trait varieties Cry1Ac and Cry2Ab) (177). Pesticidal residues from the plant were also considered but had little effect on overall conclusions. Bt cotton had less EI than conventional cotton; the EI of Cry1Ac cotton was 53% that of conventional, whereas the value for the two-trait variety was 23%. In Canada, HT canola varieties, i.e., glyphosate-, glufosinate- and imidazolinone-tolerant varieties (Section 2.6), have been cultivated on a large scale since their introduction in 1996. The EI of HT canola was determined from 1995 to 2000 using EIQ. Although HT canola acreage increased from 10% in 1996 to 80% in 2000, the AI/ha declined by 42.8% and the EI/ha, based on EIQ and amount of AI for the herbicide, declined by 36.8% (51).

A more global analysis of impacts of GE crops using EIQ was performed in 2006, comparing typical EIQ values for conventional and GE crops and aggregating these values to a national level (54). Assumptions were made to perform these calculations; e.g., pesticide use levels were based on typical herbicide and pesticide treatment regimes for conventional and GE crops provided by extension and research advisors in particular regions (258). Given the caveats of the assumptions, the conclusion was that GE crops resulted in significant reductions in the global EI of production agriculture (table 5 in Reference 54); e.g., since 1996 the overall EI associated with pesticide use on HT soybean, corn, cotton, canola, and Bt cotton decreased by 15.3%.

In 2002, under supervision of the International Union for Pure and Applied Chemistry, an international team from various fields of crop protection chemistry undertook a five-year project to analyze pesticide use in GE versus conventional crops and to estimate changes in EI (173). They used data from public sources, including the scientific literature and reports published by various institutions. In contrast to several studies prior to 2002 that focused on AI quantities and economic effects of GE crop adoption, this study estimated the EI of changes in pesticide usage (174) using 2004 data collected by NCFAP (259) on herbicide usage on GE and conventional crops in the United States. For HT canola, cotton, maize, and soybean, total quantities of herbicide AI used in general decreased from 25 to 30% compared with conventional varieties; reductions in total EI of herbicides used were also observed with GE versus conventional crops (table 1 in Reference 174). Reductions were also observed for total EI per hectare (39 to 59% reduction) and for impacts on farm workers (40 to 68% reduction), consumers (35 to 59% reduction), and ecology (39 to 55% reduction). The numbers are generalizations based on the data used and could vary among locations. Notably these results are comparable to another study (55) in which a pesticide use footprint was calculated; this study showed that the positive effects of utilizing GE varieties were greater based on EI per unit area than on AI quantities.

When looking at herbicide usage and EI, it is important to note that in addition to use on GE crops, herbicides can be applied directly to conventional crops. Also, depending on weed pressure, multiple applications can be used in the same area during the same season. Taking these facts into consideration, glyphosate use per acre has increased dramatically from 1995 to 2005, coupled with a concomitant dramatic drop in the use of other herbicides (figure 3 in Reference 174). Cultivation of GE HT crops has also had other positive effects on the environment, i.e., increases in low- or no-till practices and use in combination with integrated pest management schemes (98), which were made possible because early season pesticide sprays could be eliminated, allowing beneficial insects to establish.

Most nonanecdotal analyses on AI usage and EIQ focus on North America, mainly because most GE HT crop acreage is in this region. Recently, an analysis of the potential EI of introducing HT crops into the European Union agricultural system was undertaken

(175), despite the fact that acreage of GE crops currently in the European Union is limited. Using large-scale experimental data for HT sugar and fodder beets and to a lesser extent HT canola, it was concluded that amounts of herbicides used on HT beets were reduced, whereas those on HT soybean versus conventional were slightly higher; the latter observation is comparable to the situation in the United States. Besides North America and the European Union, other countries (e.g., Argentina, China, India, and South Africa) grow large acreages of HT and Bt varieties and pesticide usage has been studied. Most reports indicate pesticide use and cost decrease following adoption of Bt varieties (table 2 in Reference 174). In Argentina, numbers of herbicide applications increased with HT soybean but use shifted to more environmentally friendly herbicides (245).

In summary, numerous studies have been conducted on pesticide usage that analyzed different data sets and methods, sometimes leading to conflicting conclusions. Some studies showed pesticide use, expressed as AI per unit area, decreased with introduction of GE HT and Bt crops; some studies showed increases. More recently, studies have focused on EI and these have shown reductions in EI, including on farm workers, consumers, and ecology. Nonetheless, additional effort is necessary to further reduce EI of agricultural production. This goal can be achieved by using the best methods and tools available, including integrated pest management, biocontrol, organic production methods, and GE organisms (Section 2.17) to reduce EI while achieving adequate production levels.

2.8. Is It True that Bt Crops Need Additional Insecticide Applications?

Bt or Cry toxins are toxic to susceptible larvae when cleaved to generate their active form, which then binds to specific receptors in the midgut and creates holes that cause larvae to die (Section 2.1). The specificity of Cry toxins means that those aimed at lepidopteran insects (e.g., butterflies) have no effects on coleopteran insects (e.g., beetles), but this specificity is often not understood. Lack of understanding of the narrow range of Bt, compared with insecticides which control a broad range of soil pests, caused Indian and South African farmers to use fewer pesticides to control nontarget, sap-sucking pests of Bt cotton (210, 211), thus allowing secondary pests, such as grubs and cutworms, to cause damage (88).

The first Bt GE crops controlled major insect pests, such as European corn borer (*Ostrinia nubilalis*) and rootworm (*Diabrotica* spp.) for maize and bollworm (*Helicoverpa zea*) and armyworm (*Spodoptera frugiperda*, *Spodoptera exigua*) for cotton. At first only single Bt genes were used, thus minimizing collateral damage to nontarget insects, but this strategy did not completely eliminate collateral damage because some nontarget organisms belong to the same group targeted by the Bt (Section 2.1). Since pests belonging to groups insensitive to that Bt were not controlled, they were able to cause crop damage. This situation was addressed with commercial introduction of cotton varieties with two stacked Bt genes, for example, Cry1Ac and Cry2Ab, which are toxic to target bollworms and also to secondary armyworm pests (70). Although not commercialized, maize has also been engineered with six insect resistance genes against lepidopteran (Cry1F, Cry1A.105, Cry2Ab2) and rootworm (Cry34Ab1 + Cry35Ab1, modified Cry3Bb1) pests (139).

Another approach to increase the numbers of insects targeted by Cry proteins involves the use of domain swapping. Domains from one family of Bt toxins that has three domains, each with a separate role in receptor binding and channel formation, were combined to generate toxins with novel specificities. For example, a hybrid Cry protein with one domain from Cry1Ba and one from Cry1Ia conferred resistance to both lepidopteran and coleopteran pests of potato (215). Directed evolution approaches are also used to create toxins with improved receptor binding (162).

Early efforts also focused on engineering insect tolerance using plant defense proteins.

Although many efforts led to partial resistance, there were two exceptions. Genes for α-amylase inhibitors from legumes, involved in resistance to coleopteran seed weevils, were engineered into garden pea (*Pisum sativum*) and other legumes; seeds were shown to be resistant to larvae of bruchid beetles and other field pests (212). These varieties have not been commercialized in part because of possible safety concerns (241). The second exception was the introduction of lectin genes from snowdrop (*Galanthus nivalis*) into rice to control sucking insects, such as aphids and plant and leaf hoppers. Although these studies led to partial resistance to the rice brown planthopper (*Nilaparvata lugens*) (248), concerns over human safety stopped the effort [Part I of this review (186a), Section 3.2].

Other GE approaches in various stages of development involve use of insecticidal compounds from nematodes (*Heterorhabditis*), bacterial cholesterol oxidase, avidin, volatile communication compounds, and RNAi approaches targeted to specific insect proteins (see Reference 124 for a review). Even with GE approaches, other methods of insect control will be needed, e.g., chemical pesticides, biocontrol, integrated pest management, or organic approaches, because insects are plentiful and ever changing.

2.9. Can the Introduction of Virus-Resistant Genetically Engineered Plants Lead to Novel Viruses?

The first commercialized GE plant in the United States was viral-resistant squash, engineered with a viral coat protein gene (119). USDA APHIS deregulated the squash (Section 2.14), allowing commercial production after the virus was shown not to infect wild squash varieties. The resistance gene gave no advantage to wild squash varieties, and the presence of the coat protein gene did not increase viral competitiveness (176). Papaya was the second commercially cultivated plant engineered for viral resistance, also with a coat protein gene (189). Use of a coat protein gene raised concerns that another virus would infect the GE plant and, following recombination, a novel virus would arise with altered virulence, host range, or vector specificities.

This concern arose from sequence analyses of viruses, which indicated that homologous and nonhomologous recombination occurs between viruses and between viral genomes and plant genes (255). In fact, in laboratory demonstrations several viruses were shown to have viral genes in their genomes from other viruses that were introduced into the plant at the same time (118, 140, 262, 325). Experimental recombination of a transgene into a cauliflower mosaic virus (CaMV) strain, unable to infect solanaceous plants, resulted in a virus with altered symptomology and a host range that extended to some Solanaceae (262). A similar recombinational event occurred between a tomato bushy stunt tombusvirus mutant and a coat protein transgene (48).

These and other results showed that recombination does occur between transgenes and viruses and recombinants can be recovered. Functional chimeric viruses have also resulted from recombination between distinct viruses (199), proving that novel viruses do evolve under natural conditions when two or more viruses co-infect a plant. In fact, it is more likely that novel viruses would arise from cross-infection in non-GE plants because numbers of subliminal viral infections are high, thus providing ample opportunity for viral recombination. There are also fewer constraints on recombination between different viruses than between viruses and viral genes introduced into plant chromosomes (313). Experimental results indicate, however, that most recombinant viruses are not fully virulent because the new gene combinations are not fully compatible, leaving new hybrids at a competitive disadvantage (255). To compete effectively, recombinant viruses must have functional recombinational ability, capacity to establish systemic infection, and ability to compete with their progenitors during replication. These requirements place powerful negative selection pressure on newly evolved viruses. Reduced viral replication

CaMV: cauliflower mosaic virus

Selection pressure: process by which favorable traits that are inherited become more prevalent in successive generations and unfavorable traits become less common capacity could also negatively affect recombination frequency in transgenic plants.

Given the complexities of host-pathogen × environment interactions in the field, laboratory and greenhouse experiments do not provide adequate information on frequencies and fitness of novel viruses. Fitness measured in the laboratory in experimental hosts (e.g., *Nicotiana benthamiana*) does not reflect field situations where other factors, such as vector transmission, alternate host range, viral accumulation, and competition with other viruses, play a role. However, a novel virus has in fact been observed naturally in the field where no GE plants were involved; this virus resulted from the recombination of viral genes from two different strains of African cassava mosaic virus (113). Gene mixing likely took place during cross-infection by two different viral strains in the same plant.

Large-scale field releases of plants engineered with viral genes are necessary to obtain realistic assessments of the types and recombination frequencies that might occur. To date no novel viruses have been reported resulting from GE plants in the field, but likely they would be detected only if their appearance had adverse effects. At present, the only commercially propagated plants engineered with viral coat protein genes, GE squash and papaya, are grown on small acreages (164). The possibility for viruses to pick up viral coat protein genes from GE plants will be dramatically reduced in the future because strategies to create viral resistant plants will employ methods, such as RNAi-mediated viral resistance (for a review see Reference 188), that use short stretches of viral DNA, generally 300 to 800 bp (321), that do not encode proteins. For example, an RNAi construct used to silence a gene from bean golden mosaic virus in *Phaseolus vulgaris* led to virus-resistant plants (47). Such approaches minimize the possibility for gene exchange among viruses.

2.10. Can Plants and Microbes Be Engineered To Improve the Environment?

Cleaning polluted soil or water using living organisms is called bioremediation or phytoremediation when plants are used. Nonengineered bacteria and plants are able to remove heavy metals such as aluminum, selenium, mercury, and organic pollutants from contaminated soil by concentrating them in the cells of their roots, stems, or leaves (235, 257). These natural processes can be made more efficient and more directed through targeted modifications via mutation, classical breeding, or rDNA methods.

One phytoremediation effort has focused on selenium pollution, a worldwide problem arising from refinery effluents, industrial wastewater, and discharges from electric power plants. Removal was achieved in the field using fast-growing Indian mustard (*Brassica juncea*), which accumulates selenium to hundreds of parts per million (28). Other native plants hyperaccumulate selenium to thousands of parts per million and can grow in selenite-rich soils, although they accumulate little biomass (83). Engineering Indian mustard with a gene from a hyperaccumulater resulted in plants that produced greater biomass and longer roots, and accumulation and volatilization of selenium was significantly increased (184). In field experiments, engineered Indian mustard plants, overexpressing adenosine triphosphate sulfurylase, gamma-glutamyl-cysteine synthetase, and glutathione synthetase, were shown to contain approximately three- to four-fold more selenium in their leaves than wild-type plants (27).

Mercury is one of the most hazardous heavy metals and is particularly problematic in aquatic environments where organic mercury moves from fish to humans. *Arabidopsis thaliana* engineered to express modified bacterial mercuric ion reductase detoxified mercury by converting the more toxic ionic form to a less toxic elemental form, Hg(0) (46). To address mercury pollution in riparian ecosystems, Eastern cottonwood (*Populus deltoides*) engineered with the same gene was shown to evolve two- to four-fold the amount of Hg(0) and accumulate significantly higher biomass compared to control plants in soils contaminated with 40 ppm of ionic mercury, demonstrating the potential for in situ mercury remediation from soils (68). Concerns have been raised that this strategy might move pollution to the atmosphere, from which it would be redeposited onto land (45).

Another potential environmental advantage of GE plants is making better use of resources such as land and water. This goal can be achieved by engineering plants (*a*) to achieve higher yields using the same levels of inputs, e.g., 12% increase in yield by engineering rice with the *Zea mays* phosphoenolpyruvate carboxylase gene (181); (*b*) to incur fewer losses due to pests, by engineering potato (*Solanum tuberosum*) against late blight (*Phytophthera infestans*) disease (278); and (*c*) to survive in soils with high concentrations of salt (336) or lower levels of water (265). Lowering crop production losses reduces the likelihood that environmentally sensitive areas will be cultivated, potentially allowing maintenance or even expansion of protected forests, lakes, shores, wetlands, and wilderness.

2.11. Can Genes From Genetically Engineered Plants Move to Bacteria in the Field?

Transfer of genes among nonsexually related organisms, e.g., from plants to bacteria, is called horizontal gene transfer. It can occur in nature among sexually incompatible bacteria and may have played an important role in bacterial evolution (117). Horizontal transfer is in contrast to vertical gene transfer, where an organism receives genetic material from its parent or a species from which it evolved. Recent sequence analyses of genes and proteins show that some genes have transferred from plants to bacteria (42, 90); however, this exchange occurred over a very long evolutionary timeframe. Many factors limit frequency of transfer, especially between kingdoms such as plants and bacteria (42). The only successful recent demonstration of plant to bacterium transfer of DNA has occurred under optimized laboratory conditions—situations difficult to replicate in natural settings (125). Numerous field studies have failed to show horizontal transfer at detectable frequencies between plants and bacteria (53, 260). Thus, if such an event were to happen in the field, it would be at very low frequencies and the gene would need to provide a selective advantage to survive during generation advance.

Predictions regarding horizontal transfer were tested using DNA from sugar beet engineered with a kanamycin resistance (Km^r) gene. Total DNA from GE sugar beet was spiked into sterile soil, to which were added nutrients and the bacterium *Acinetobacter* harboring a neomycin phosphotransferase II (*nptII*) gene with a 317 bp deletion that caused it to be kanamycin sensitive (Km^s) (227). Noncompetent bacteria integrated a fragment from plant DNA that restored Km^r at a frequency of 2.2×10^{-8}, but only in sterile soils, a situation that would be unlikely to occur in nature. On the basis of earlier studies, recombination frequencies in nonsterile soils were 10^{-10} to 10^{-11} (126, 226), and frequencies were further reduced in either soil type if no homology existed between donor and recipient DNA.

Effects of selection pressure on persistence of Km^r bacteria were assessed by adding increasing levels of Km to the soil. This led to the conclusion that natural soil conditions rarely would have the selective pressure necessary to keep *nptII* in the bacterium (227). Data from this and other studies indicate that homologous recombination and integration of plant genes into competent soil bacteria could occur, but at very low frequencies, and the environmental significance would depend on selective pressure for the trait. Thus, the nature of the gene, whether naturally occurring or GE, would dictate risk.

Km^r: kanamycin resistant

npt: neomycin phosphotransferase

2.12. Is the Loss of Honeybees Due to Genetically Engineered Crops?

"Bees Vanish, and Scientists Race for Reasons," quotes the New York Times in April 2007 (29). Readers might worry about honeybees (*Apis melliferea* L.) because of the honey they produce, but the greater effect would be because they would not be available to pollinate almost 90 different fruit, vegetable, and crop species in the United States (76). In the latter case, a lack

CCD: colony collapse disorder

FDA: Food and Drug Administration

of adequate honeybee populations could have serious consequences.

Honeybee die-offs had occurred before, e.g., in 1998 with the introduction of varroa mites into the United States, which resulted in declines in honeybee colonies from 80,000 in 1982 to 38,500 in 2004. But in spring 2007, colony die-offs with new symptoms, termed colony collapse disorder (CCD), occurred in several European Union countries, the United States, and Canada (167).

A connection between Bt maize and CCD was raised in experiments conducted in Germany that were described on the Internet but never published in a scientific journal (128). In these studies honeybees were fed Bt maize pollen and, although healthy bees had no acute or chronic toxic symptoms, in one experiment where bees were infested with parasites, the study was aborted because Bt pollen appeared to accelerate the bees' decline. Although not repeatable in subsequent experiments, Bt in GE corn pollen thus became a possible cause of CCD.

Prior to these experiments, however, numerous studies had determined the impacts of Bt on bees: (*a*) Canadian scientists found no effects of pollen from Bt sweet corn on honeybee mortality (24); (*b*) Mexican scientists found no effects of different syrups with Cry1Ab protein on bee colonies (247); (*c*) exposing bees to 1000 times more Cry3b than in pollen resulted in no toxic effects on bee larvae or pupal weight (15); and (*d*) feeding honeybees pollen from Cry1Ab maize did not affect survival, gut flora, or development of hypopharyngeal glands, where protein-rich food for the brood is produced (20, 22, 23). In 2008 a meta-analysis of 25 independent studies assessing effects of Bt Cry proteins on honeybee survival (mortality) showed that Bt proteins used in commercialized GE crops to control lepidopteran and coleopteran pests do not negatively impact the survival of honeybee larvae or adults (91).

Thus there are no data in the scientific literature supporting direct or indirect damage to bees caused by currently approved GE crops engineered to make Bt proteins. Additionally, larvae consume only a small percent of their protein from pollen (21, 23), and there is also a lack of geographic correlation between GE crop locations and regions where CCD occurs. For example, CCD was reported in Switzerland, where no GE crops are grown (167). Other causes have been suggested, such as exposure to chemicals, pesticides and other stress factors; lack of genetic diversity in honeybees; and immune suppression (67). Several pathogens have also been implicated, e.g., a spore-forming parasite, *Nosema ceranae* (297), and the Israeli acute paralysis virus (77).

2.13. Can Federal Regulatory Agencies Stop Planting of Genetically Engineered Crops That Pose Environmental Risks?

The United States created a formal regulatory structure for GE organisms establishing the concept that GE foods would be regulated on the basis of product, not process, and would be regulated on a case-by-case basis (229) [Part I, Section 2.6 (186a)]. GE crops and products made from them are under regulatory control of three federal agencies: the Food and Drug Administration (FDA), the EPA, and the USDA (for a review, see Reference 201). The FDA is responsible for food safety and labeling of foods and animal feeds from conventional and GE crops. The EPA evaluates food safety and environmental issues associated with new pesticides and pesticidal products, such as Bt corn and the pesticidal Bt product it contains. The EPA's charge also includes GE plants in which a small part of a pest, such as a viral regulatory sequence (e.g., 35S promoter), is used. A division of the USDA, APHIS, oversees environmental safety of planting and field-testing GE plants to ensure GE crop field tests are performed under specified conditions and any unusual occurrences are reported. All three agencies do not oversee each GE crop; however, all have legal rights to demand immediate market removal of any product if valid scientific data show safety concerns for consumers or the environment.

A plant with an rDNA fragment inserted into the plant genome is considered a regulated article by USDA APHIS, and each time a specific fragment is inserted it is considered a new event [Part I, Section 2.6 (186a)]. Each event must go through regulatory approval, even if a first event with the same fragment received approval. Regulated articles are evaluated for impact on the environment and on agriculture, e.g., will the gene move to a native plant and perturb the ecosystem or become a weed in a cultivated setting? Small-scale field trials are used to make preliminary EI assessments. In the United States, the GE plant is a regulated article until APHIS deregulates it [Part I, Section 2.6 (186a)]. To gain nonregulated status, molecular, biochemical, and cellular analyses are done on the GE plant, and data are collected on the life cycle, reproductive characteristics, and expected and unexpected changes versus a nonengineered plant. A petition for nonregulated status containing these data is formulated by the event's creator and reviewed by APHIS, after which an Environmental Assessment can be issued and determination of nonregulated status granted. From June 1992 to January 2009, 117 petitions for nonregulation were received at APHIS. Twenty-nine were withdrawn or are incomplete; 13 are pending; 75 petitions have received nonregulated status (160), including GE varieties of chicory, corn, cotton, flax, papaya, plum, potato, rapeseed, rice, soybean, squash, sugar beet, tobacco, and tomato. Information on status, requesting institution, genes introduced, phenotype of GE plants, field test data, and environmental assessments of deregulated articles is publicly available (160). Deregulation does not mean the GE crop has been commercialized, only that it no longer requires APHIS review for movement or release (161).

In 2005, the USDA Inspector General conducted an audit that indicated the USDA lacked basic information about where GE crops were grown and their fate after harvest (155). This finding raised concerns, particularly about crops that produce pharmaceuticals. Although all three federal agencies can legally request removal of a product from the market, it was the court system that made inquiries regarding the EI of two GE crops, one which had nonregulated status and one which had not yet requested such status. In the first instance, a U.S. federal court ordered the USDA to conduct more detailed reviews of applications for experimental plots of GE bentgrass after it was shown that pollen had spread thirteen miles from the original cultivation site (320) (Section 2.14). The second instance involved Roundup Ready® alfalfa. In 2005 APHIS concluded that this GE variety was safe for animal feed on the basis of substantial equivalence; ~320,000 acres (129,500 hectares) were subsequently planted in the United States. A U.S. District Court Judge for the Northern District of California, however, ruled that the USDA had erred in approving deregulation (9) and that nonregulation (Section 2.14) might have significant EI that required preparation of an environmental impact statement (EIS). The court further stated that the USDA violated the National Environmental Policy Act by preparing an environmental assessment (EA) instead of an EIS (11). After the court ruling in March 2007, further plantings of HT alfalfa were prohibited and restrictions were put on its production. Roundup Ready® alfalfa returned to regulated status, pending submission and review of an EIS (11, 13).

Substantial equivalence: used to determine whether a new food shares similar health and nutritional characteristics with existing, familiar foods with demonstrated histories of safe use

EIS: environmental impact statement

EA: environmental assessment

Adventitious presence (AP): technically unavoidable, unintended presence of undesired material in an agricultural commodity

2.14. What Happens When Pollen Moves From Genetically Engineered Crops to Wild Relatives or Non-Genetically Engineered Varieties? In Areas of Genetic Diversity?

Most plants reproduce via self-fertilization or movement of genes from one parent to another via pollen. In fact, this process is an essential tenet of genetic diversity. But movement of unwanted genes, naturally occurring or engineered, may result in adventitious presence (AP), a situation where unwanted substances unavoidably are present in production and marketing of agricultural products. AP can occur for a variety of reasons, including gene flow, and sometimes results in economic consequences for commercial GE crops (75).

Generalizations about whether gene flow presents significant economic or environmental risks cannot be made for either conventionally bred or GE crops; case-by-case evaluation is required. Many major agricultural crops are sexually compatible with wild and/or weedy relatives, and, if the plants grow in overlapping regions, crop-to-weed or crop-to-wild relative gene flow could result (16; for a review see Reference 96). This outcrossing to wild populations can result in new combinations of genes that can improve, harm, or have no effect on the fitness of recipient plants. Genes can also flow from wild relatives to cultivated crops, introducing new traits into next generation seed, but only affect the crop if it is replanted. Gene transfer among plants may be a larger containment issue than unwanted pesticides, because genes reproduce in the recipient plant (95).

Pollen drift is a major, although not the only, conduit through which unwanted genes end up in crops. Numerous factors affect the frequency of gene flow resulting from pollen drift, i.e., biology of the species, the environment, and production practices, and these should be considered in developing strategies to minimize gene flow. Successful cross-pollination requires that parental plants (*a*) flower at the same time; (*b*) be close enough to allow a vector (insect, wind, or animal) to transfer pollen to receptive females; and (*c*) produce pollen that can result in embryos developing into viable seeds and germinating (for a review see Reference 194). Successful pollination also depends on the longevity of pollen viability and the distance it must travel (96, 97). Also important is whether the plant self-pollinates, as is the case for tomatoes, soybeans, and most cereal crops, or is open-pollinated, as in the case for corn and canola, where pollen from one plant fertilizes another. Gene flow is more frequent with the latter.

There are wild, weedy species in the United States compatible with some existing or anticipated commercialized GE crops. The first GE trait in a commercial crop with wild relatives in the United States was virus-resistant squash (157). USDA APHIS determined the impact of gene flow of the GE trait to wild varieties; the squash received nonregulated status (Section 2.13) and was grown commercially after it was shown that viruses against which resistance was directed did not infect wild varieties or increase their competitiveness (176).

HT traits have been engineered into major U.S. commercial crops such as canola, corn, cotton, and soybean. Whether gene flow of HT traits leads to more competitive, herbicide-resistant weeds depends on factors such as species, location, and trait. One crop for which this might be a concern in the United States is cultivated rice, which outcrosses with perennial, wild red rice (*Oryza rufipogon* Griff.), considered a noxious weed in the United States. Red rice is sexually compatible with cultivated rice, grows in many of the same regions, often has overlapping flowering times, and thus is a prime candidate for gene flow with cultivated rice. Breeders generally try to avoid gene movement from red rice to cultivated varieties because of its undesirable traits, e.g., awned seeds and red pericarp. When GE HT traits were introduced into cultivated rice, attention shifted to the impact of genes moving into red rice. To study this, experiments were conducted to determine gene flow rates under natural field conditions from cultivated rice to wild red rice and weedy rice (*O. sativa* f. *spontanea*) in China and Korea, respectively (69). An HT gene and simple sequence repeat (SSR) fingerprinting were used to monitor gene flow, which ranged from 0.01 to 0.05% for weedy rice and from 1.21 to 2.19% for wild red rice. Although frequencies were low, gene flow did occur, emphasizing the need to avoid outcrossing when genes could enhance the ecological fitness of weedy species. In another study, resistance to imidazolinone herbicides, created by mutagenesis, not by engineering, was used to assess gene flow and fitness of the recipient (79), a reminder that gene flow is not limited to GE varieties and its impact is dependent on the trait, not the means by which the gene was created.

Numerous studies have evaluated pollen-mediated, intraspecies gene flow from canola to its wild relatives. One study evaluated the outcrossing of *B. napus* with wild relatives, including *B. rapa* L. (rapeseed), *Raphanus raphanistrum* L., *Sinapis arvensis* L., and *Erucastrum gallicum* (318). Hybridization between *B. napus* and *B. rapa* in two field experiments was ~7% in commercial fields and ~13.6% in the wild. Gene flow from GE *B. napus* to the other three wild varieties was shown to be low (<2 to 5×10^{-5}); however, genes could move into the environment via wild *B. rapa* or commercial *B. rapa* volunteers. Analysis of 16 of these types of studies identified major factors affecting pollen-mediated gene flow from *B. napus* (152), using either a donor plot surrounded by receptor plants (continuous design) or a receptor field only on one side of the donor plot (discontinuous design). With continuous designs, cross-fertilization averaged 1.78% ± 2.48% immediately adjacent to the donor plot and was fairly constant at 0.05% ± 0.05% at distances over ten meters (1 meter = 3.3 feet). With discontinuous designs, outcrossing rates were 0.94% (±0.51) next to donors and 0.1% (±0.11) at distances over 100 meters. Thus, most outcrossing occurred in the first ten meters from the field, although numerous factors relating to the field, plant, pollen, and environment influenced the rate. Aside from pollen flow, volunteer HT populations can also arise via seed-mediated flow (143) and from feral populations (152).

An example of a trait moving from a commercial GE crop to a non-GE variety is triple-resistant canola (146) (Section 2.3); this outcome was predicted prior to release of the GE variety because of canola's tendency to outcross (36). The multiply resistant volunteers, with two GE traits and one mutant HT trait, could still be controlled with other herbicides; however, their presence has decreased the utility of HT canola (66). Movement of HT genes could have been monitored more closely to prolong the effectiveness of the HT varieties.

A well-publicized study of pollen-mediated gene flow involved precommercial GE HT creeping bentgrass (*Agrostis stolonifera* L.), a wind-pollinated, highly outcrossing, perennial grass (320) (Section 2.13). Because bentgrass has native, weedy relatives in the United States with which it outcrosses (39), transgene movement to related *Agrostis* species and dissemination of seeds and vegetative propagules were examined. Following a single growing season of GE bentgrass, most transgene flow was found within 1.2 mi (2 km) in the direction of prevailing winds; limited gene flow was found to 13 mi (21 km). Further study showed that nine HT creeping bentgrass plants (0.04% of samples) grew ~2.4 mi (3.8 km) beyond the control area—six from pollen-mediated gene flow in the direction of prevailing winds and three from dispersed GE seeds (250). Three years after production halted in HT bentgrass fields, 62% of 585 bentgrass plants tested positive for the HT gene; 0.012% of seedlings from seed of HT plants were HT positive (335), suggesting that under some conditions transgenes can establish in wild populations after short exposures. Although no long-term ecological studies were done, it was suggested that herbicide application or drift could lead to persistence of the HT trait in wild plants (249).

Prior to commercial release of HT alfalfa (Section 2.13), studies were done to assess gene flow in fields grown for seed and for forage (for review, see Reference 310). Under intentionally poorly managed fields (20 to 50% bloom), gene flow from a forage field to a seed field was <0.5% at 165 ft and 0.01% at 350 to 600 ft (289). This type of information can be used to establish distances and practices to minimize gene flow (310). Gene flow also occurs to feral alfalfa, frequently found growing outside cultivation areas, and this type of gene flow is affected by the same barriers as other alfalfa gene flow, i.e., flowering synchrony, presence of pollinators, and distances between alfalfa fields and feral plants. Gene flow to feral alfalfa, which is less abundant and less conducive to seed set, can be reduced by decreasing feral flowers through frequent mowing or animal predation.

In areas of genetic diversity of plants related to GE varieties, additional precautions are

NOP: National Organic Program

needed to reduce possible impacts of introgression of GE traits, when potential significant environmental consequences could occur, and to minimize this occurrence. For example, outcrossing of GE HT rice with wild rice varieties has potentially significant environmental such impacts, whereas gene flow of the vitamin A trait from GE Golden Rice is less likely to have such impacts. Where possible impact is significant, planting of GE crops near wild species should be avoided or GURT-type technologies could be used to prevent gene(s) from moving to wild varieties (Section 3.2).

On the basis of published studies, gene flow will occur when compatible plants are present and thus GE traits can move and persist in unintended plants. Even in the absence of gene flow, GE varieties can persist in the agricultural environment. For example, in Sweden, GE volunteer oilseed rape plants (0.01 plant per m^2) were observed ten years after a trial of GE HT oilseed rape (84). Farmers need to be cognizant of gene movement from GE crops and the possible persistence of GE varieties. For organic farmers the presence of GE traits in their crops could create a problem if a contract was signed limiting the presence of GE traits in their organic products (Section 2.15). Conventional farmers should also be aware of transgene movement to a non-GE crop if it is intended for export or other sensitive markets (Section 3.4).

2.15. What Happens When Pollen Moves from Genetically Engineered Crops to Organic Crops?

Organic farming is a production system in which, among other restrictions, synthetically produced fertilizers and pesticides are not permitted; control of biotic pests is accomplished by biological pest control and nonsynthetic pesticides such as copper, rotenone, and Bt (252). Although some GE crops are engineered to produce Bt, use of GE crop varieties in certified organic farming is specifically prohibited (252). To be sold or labeled as 100% organic, the product must be produced and handled without use of excluded methods that include "... recombinant DNA technology (including gene deletion, gene doubling, introducing a foreign gene and changing the positions of genes when achieved by recombinant DNA technology)." Excluded methods "do not include traditional breeding, conjugation, fermentation, hybridization, in vitro fertilization or tissue culture" (224). Despite this ban on GE technology, some argue that GE crops could fill a niche in organic farming (253).

In the United States, organic production is a process, not a product certification, and thus does not specify the nature of the food or ingredient. Although organic farmers are not required to test for pesticides (4), AP of certain excluded materials, such as synthetic pesticides, are permitted (5). Presence in an organic product of a particular pesticide at levels <5% of the EPA's tolerances can be labeled and sold as organically produced (221). Presently there is no policy on acceptable thresholds for the unintended presence of GE materials in organic foods or products. Problems of gene flow to organic fields are similar in some ways to pesticide drift to organic farms from aerial spraying. The USDA set specific limits for pesticide presence and minimal distances between fields and a similar approach could be developed for GE crops if zero tolerance is not the goal (Section 2.16).

Because of the ban on GE crops in organic farming, some believe an organic farmer will automatically lose his/her accreditation if the crop is unintentionally mixed with a GE crop (252). The presence of detectable levels of GE material in a crop does not constitute a violation of National Organic Program (NOP) regulations nor is it reason to lose accreditation, as long as the grower has not intentionally planted GE seed and has taken reasonable steps to avoid cross pollination (252; for specific wording of NOP standards, see Reference 220). The USDA-NOP informed state agricultural departments that up to 2005 no organic farmer had lost organic certification because of AP of GE material (172, 291). However, the organic farmer might lose income from GE presence, if the product is being provided under a personal contract guaranteeing a 100%

GE-free product. This is not an NOP rule but a private agreement between grower and buyer (171).

Some consumers, however, expect foods labeled as organic not to contain GE ingredients and have zero tolerance for their presence. Achieving 100% purity for any agricultural commodity is a practical impossibility given the nature of our food system, the reproductive biology of plants, and the highly sensitive detection methods available to identify GE traits (270). These latter methods include PCR assays, which require knowledge of the DNA sequence introduced (78), and enzyme-linked immunosorbent assays (ELISA), based on antibodies specific for the introduced protein. These testing methods establish GE presence and can result in extra costs to the producer; however, not conducting such tests could mean also extra costs because of rejection at the point of sales.

2.16. Can Organic, Conventional and Genetically Engineered Cropping Systems Coexist?

The coexistence of differing varieties and production methods is not new to agriculture. Breeders and farmers have developed strategies to grow and market different varieties, such as white and yellow maize, hot and sweet peppers, high- and zero-erucic acid rapeseed, and still achieve purity standards dictated by certified seed specifications. When producing crops in or for countries where labeling thresholds exist for AP of GE products (Section 2.14), methods must be established to separate different product lines to enable coexistence so the economic needs of all farmers can be met (156) (Section 3.4).

Farmers also have to choose among various production methods to grow their crops; it is not uncommon for different farming systems to be used on adjoining fields. Thus, farmers have to deal with mixing of permissible inputs and methods, whether within their own farms, with products from neighboring farms, or during harvest and processing. This commingling or AP is the unintended occurrence of materials other than the specific crop and can include weed seeds, seeds from other crops, dirt, insects, and other foreign material, such as stones or plastic. For seed crops, rules for AP are specified by the Association of Official Seed Certifying Agencies (AOSCA). For example, a level of 0.5% seed of other varieties and 2% AP of inert material is permitted in "pure seed" of hybrid corn seed (172).

Historically, dealing with practices of neighboring farms has been handled by farmers working with each other to minimize impacts. This situation occurs, for example, when synthetic pesticides are used on conventional farms and organic farming is being practiced on adjoining fields. This situation can cause economic losses for the organic farmer when prohibited pesticide residues (222) occur at levels >5% of the EPA's tolerances, because the product cannot be sold, labeled, or represented as organically produced (223). Thus, coexistence strategies must be devised to allow both neighbors to farm in an economically viable manner. This can involve alerting each other to their plans and modifying them to accommodate each others' needs. When GE crops are grown next to organic farming operations, certain practices that minimize synthetic pesticide drift can also limit GE gene flow, such as spatial separation of fields, staggered planting dates, and planting varieties with different maturity dates and those that are not sexually compatible. Other crop-specific methods have been devised to aid coexistence strategies (52, 153, 244). Gene flow is not the only means for GE to commingle with conventional or organic crops; crops must also be segregated during harvest, shipping, and processing. Methods limiting such commingling have in some cases been implemented (52, 59, 153, 233, 244).

The European Commission on Agriculture and Rural Development adopted guidelines in 2003 for the development of national strategies and best practices in the European Union to ensure coexistence of GE crops with conventional and organic farming (72). Individual European Union countries have developed their own

ELISA: enzyme-linked immunosorbent assay

AOSCA: Association of Official Seed Certifying Agencies coexistence strategies (129), which has led to differences in the legal and economic situations within the European Union (37). Country-to-country growing conditions are so varied and experience with GE crops is still so limited that it is difficult at present to develop unified legislation on coexistence. The European Commission is set to release a report describing the development of national coexistence measures (130).

In 2006 the UK DEFRA outlined a protocol on how to manage coexistence, with stricter standards for coexistence with organic products (86). The aim was to ensure future growing of GE crops without resultant disadvantages to any farmer. The legislation establishes statutory separation distances between compatible GE and non-GE crops, which are specific for each crop and which account for the size of receptor and donor fields, and a statutory notification process to inform neighboring farms. Nonstatutory recommendations include control of volunteers and bolters (sugar beet) and cleaning of shared combine harvesters. Feedback was sought from stakeholders (130) with the intent that by the time GE crops are released in the United Kingdom appropriate coexistence measures will be in place. To facilitate spread of information and compliance, a web-based Coexistence Information System was created to share information on studies being conducted in individual European Union countries on specific GE crops (131).

One factor hampering coexistence is the demand for zero tolerance for GE presence. Achieving 100% purity with any biological system is impossible and would require a complete ban on growing GE crops. In the United States AOSCA sets "minimum standards for genetic purity and identity and recommended minimum standards for seed quality for the different classes of certified seed" (17), and this is made possible through coordinated efforts of official seed certification agencies that evaluate, document, and verify that a seed or plant product meets accepted standards (49). Mandatory practices are established by state affiliates of AOSCA that set isolation distances from fields of the same crop, use of buffer rows, and specified agronomic practices, such as rouging of undesirable plants, weed control, and detasseling (60). This type of approach could also assure reasonable purity standards for commingling of GE in agricultural products. When UK officials drafted DEFRA guidelines, they recognized that rules need to be achievable because the more complex the system, the more likely farmers would be to err or ignore the rules (34). Also critical is establishment of accurate methods to test for GE presence, availability of a testing facility, an economical cost for testing, established liability criteria, and compensation schemes once GE presence is detected (202).

2.17. Can Use of Genetically Engineered Crops or Organic Farming Lead to More Sustainable Agricultural Production Systems?

Sustainability has no single meaning, but one accepted definition is to meet the basic needs of today's inhabitants while preserving resources to enable future generations to flourish. Sustainability has become a goal of the United Nations' Development Group's Millennium project, "to 'Ensure Environmental Sustainability' by integrating principles of sustainable development into a country's policies and programs to reverse the loss of environmental resources" (295). Although the need for sustainable agricultural systems is now widely accepted, the manner in which to achieve them is not universal and even the precise goals are not well defined.

The prevailing agricultural system in the United States, so-called conventional farming, has led to impressive gains in productivity and efficiency. Some estimates are that between 70 and 90% of recent increases in food production resulted from changes in conventional agricultural practices rather than cultivation acreage increases (132). This high production does have negative environmental impacts, as well as sizeable consumption of fossil fuels, unsustainable rates of water use and topsoil loss, and contributions to environmental degradation, e.g., air pollution, soil erosion, reduced biodiversity, pest resistance, pollution of lakes and streams, and overuse of surface and ground water (149).

To achieve agricultural sustainability, causes and cures for these problems must be addressed through all possible means. Numerous agricultural practices or methods, such as integrated pest management (IPM), biological control, organic methods, and use of GE plants, coupled with selected conventional agricultural methods, can play important roles in future sustainable agricultural practices. For example, practitioners of integrated pest management use comprehensive information on the life cycles of pests and their interactions with the environment, in combination with available pest control methods, to manage pest damage with the least possible hazard to people, property, and the environment (104). Biological control involves the use of a specific living organism to control a particular pest and cause the least harm to beneficial insects (228). USDA APHIS, for example, recently released a finding of no significant impact relative to the environmental release of gall wasp (*Aulacidea acroptilonica*) for biological control of Russian knapweed (*Acroptilon repens*) (12). Biological control can be a part of an IPM strategy and neither biological control nor IPM specifically excludes the use of GE organisms.

Organic production (Section 2.15) relies on practices, such as cultural and biological pest management, that can include IPM and biological control but excludes the use of synthetic chemicals and GE organisms (300). The use of GE organisms can also contribute to sustainable practices by augmenting and replacing certain conventional practices. For example, plants can be created that increase water use (251) and fertilizer (271) efficiencies, that remediate soil contaminants (183), increase no-till or low-till practices (280) to help reduce greenhouse gases (92), and produce higher yields without increasing land usage, particularly in developing countries (41, 246). Although GE plants can contribute to a more sustainable agriculture, their development and availability do not ensure positive contributions. That depends on how they are deployed and whether their use results in changes in farming practices that increase sustainability. To achieve true sustainability agriculture must use the best of all practices.

IPM: integrated pest management

3. SOCIOECONOMIC ISSUES

When considering effects of GE crops and products, just as with those produced by other agricultural methods, it is important to factor in economic and social implications. In this section, consideration is given to impacts of GE crops on farmers and their practices, to the mechanisms of agricultural change, and to effects on developing countries. Not all issues of interest are discussed. Perhaps more difficult in these subject areas is the fact that there are not always definitive, factual responses to the issues raised; responses often reflect attitudes rather than information based on peer-reviewed scientific literature.

3.1. Why Do Farmers Plant Genetically Engineered Crops and Who Profits From Them?

Whether measured as crop yield per acre or average output per farm worker, U.S. agricultural productivity is among the highest in the world and it has increased over time. In 2004, total agricultural productivity was 2.7 times higher than it was in 1948 (120). Nonetheless, farming is at best a low profit margin endeavor, and profitability often depends on factors outside the farmer's control, e.g., weather, pest infestation, and market fluctuations. However, expected profitability plays a large role in decisions by farmers to adopt new innovations.

GE varieties can have potential positive economic impacts, but certain factors should be kept in mind. (*a*) The nature and performance of GE varieties change over time and in different locations. (*b*) No single method of assessing net economic impact of new crops is sufficient to accurately predict outcomes. (*c*) The length of time over which particular varieties are used influences assessments (273). Economic studies should also take into account impacts on labor,

ERS: Economic Research Service

Mycotoxins: toxic secondary metabolites, such as aflatoxin and fumonisin, produced by certain species of fungi or molds

health, environment, equity, and poverty. Consideration of all these latter factors distinguishes GE crops from other modern varieties because risk assessments and the potential impacts of GE crops relative to these factors play larger roles in acceptance of GE crops than for those created by traditional practices.

One important factor for farmers in considering crop profitability is yield. Although current GE crops are not engineered for higher yield per se, increased yields have been observed. This higher yield has been demonstrated in numerous studies and surveys of HT corn, Bt cotton, and Bt corn (table 3 in Reference 108). Data analysis of the USDA Economic Research Service's (ERS) Agricultural and Resource Management Surveys of 2001 to 2003 showed that most farmers, e.g., 79% of those choosing Bt corn, adopted GE varieties to increase yields through improved pest control (figure 7 in Reference 108). Other reasons included time savings and ease of agricultural practices.

In determining the profitability of Bt corn engineered for European corn borer protection, it is important to note that farmers must decide whether to purchase the more expensive Bt corn seeds before they know what the extent of insect damage to their crop will be. In years when corn borer infestations are high, farmers make a profit primarily because of increased yields. When insect pressure is low, yield losses to insect damage are slight and seed costs exceed profits (182, 326). Aside from yield considerations, another economic benefit of Bt corn is reduction of mycotoxins present in grain because of infection by toxin-producing fungi. The most prevalent impacts are due to aflatoxin, with lesser effects from *Fusarium* mycotoxins, or fumonisins, and deoxynivalenol (DON) also called vomitoxin because it induces vomiting and hemolysis of erythrocytes in animals. These compounds are known to cause a variety of short- and long-term health effects. Bt reduces insect damage on kernels, thus reducing infection by mycotoxigenic fungi (25). Economic losses are due to market rejection of contaminated grain, export market losses, and testing costs. A literature review in 2007 concluded that economic benefits of Bt maize in reducing the mycotoxins, fumonisin and aflatoxin, were ∼\$22 M and \$14 M, respectively (331). Mycotoxins are a significant health issue where unprocessed corn is a dietary staple (332), and thus, health benefits from mycotoxin reduction are particularly important in developing countries. When considering exports to these countries, the health situation could be improved by stricter mycotoxin standards; however, these standards would have negative economic impacts on major corn-exporting countries, i.e., the United States, China, and Argentina (331).

In the European Union, GE crops are planted on a limited area. Of European Union member countries, Spain grew the largest acreage (250,000 acres, 0.1 million hectares) in 2007 (165). In fact, Spain has grown commercial Bt maize for more than nine years; 15% of their total acreage is composed of Bt varieties and in regions with high corn borer infestation it can reach 60% (133). Economic analyses were performed using data from face-to-face surveys with Spanish farmers in the three leading Bt corn-growing regions that accounted for ∼90% of cultivated GE corn in 2006. A statistically significant ($P < 0.001$) 11.8% yield increase was observed in one region, Zaragoza, during three growing seasons, with lesser increases observed in the other two regions. Yield variation in these latter areas could be due to use of unadapted Bt varieties and to variations in pest pressure, but it is not due to Bt resistance development in corn borer populations (133). In one region, total revenues minus variable costs for Bt farmers versus conventional farmers were as high as ∼\$69 per acre per year higher, which compensates for the price premium on seeds. Similar yield advantages were observed in South Africa (137). The Spanish surveys revealed that most farmers adopted Bt corn to lower corn borer damage; the main reason for not adopting was reluctance to change.

Seven other European Union countries grow smaller acreages of GE crops than Spain: the Czech Republic, France, Portugal, Germany, Slovakia, Romania, and Poland

(165). In 2007 the Czech Republic grew ~1.23 million acres (0.5 million hectares) of GE maize; additional income for Bt maize in the many areas of high infestation was as high as 2430 Czech koruny ($145) per acre (123). Similar analyses for HT sugar beet showed that, taking into account treatment of HT sugar beet and additional seed costs, farmers could still achieve a 1620 koruny ($96) additional profit per acre.

Studies on economic impacts on farmers in developing countries have also been conducted. One study in India showed increases in yield and revenue with Bt cotton compared with non-Bt cotton using farmer plot rather than trial plot data, although there was some variation among subregions (211) and a few areas did not benefit (41). Yield increases in India improved when coupled with IPM practices (Section 2.17) (26). A study of farm-level preproduction trials in China showed that compared with households cultivating non-GE rice, small and poor-farm households, without the aid of experimental station technicians, realized both higher crop yields and reduced pesticide use after adopting GE rice varieties (150). In some studies, farmers in developing countries realized greater yield benefits from such crops than in developed countries. It was suggested that this result was caused by small-scale farmers suffering larger pest-related yield losses because they do not have the technical or economic resources to manage pest infestations (41, 246). To realize the greatest economic benefits in developing countries, it is important, when selecting GE targets, to consider local production conditions, consumption preferences, appropriateness of local varieties, adequacy of biosafety regulatory policies, and possible impacts of marketing issues and consumer attitudes (94).

3.2. Will Plants with Terminator-Type Genes Prevent Replanting of Genetically Engineered Crops?

The Rural Advancement Foundation International [RAFI, now ETC (Erosion, Technology, and Conservation) Group] (256) first used the term "terminator technology" in 1998 in a patent issued jointly to Delta & Pine Land Company and the USDA (315). This technology was described as a means to restrict reuse of GE seeds; second-generation seeds would be sterile (230) and could not germinate (144).

Terminator technology is one form of GURT; there are two types, V-GURTs (variety-protected GURTs) and T-GURTs (trait-specific GURTs). An example of plants with V-GURTs would be those with terminator technology. Because saved seeds would not germinate, users would have to repurchase seeds each year—similar to the situation with hybrid crops that must be purchased yearly to realize yield advantages (115). Hybrid seed use, which represented 95% of U.S. corn acreage in 2006 (309), would not be affected by the use of terminator technology and users' having to repurchase seeds because farmers using hybrid seed must already repurchase seed each year. Crops engineered with T-GURTs must be treated with specific chemicals for the engineered trait to be expressed. In this case, farmers could replant seed but would lose the advantage of the trait if their crop was not treated with the chemical, something RAFI termed "traitor technology."

Terminator technology was complex as patented, involving several genes, with one stopping protein synthesis and preventing seed germination. So that the planted seed could initially grow, this gene product was not expressed in the first generation, but was instead halted by a spacer gene under control of the *cre/lox* system. Cre recombinase excised the spacer, straddled by *lox* excision signal sequences, which activated expression of a second gene, which halted germination and encoded a ribosomal inhibitor protein (RIP), under control of a third gene product for the TN10 tetracycline repressor. Use in tobacco and cotton was described in the patent as functional but efficacy was not shown. The system required functional and timed expression of three genes, making it problematic as a commercial approach, and in fact, the system has not been commercialized.

Terminator technology has been criticized by some farmer and consumer groups as potentially disastrous for food security and biodiversity (266). After criticisms surfaced in 1999, Monsanto, then owner of the technology, vowed not to use it; other seed companies also agreed. Controversy over the technology reignited in 2005 because of a statement in Monsanto's Pledge Report to Stakeholders, "Monsanto does not rule out the potential development and use of one of these technologies in the future" (208). This controversy led to the introduction of a bill into the Canadian Parliament in May 2007 to "prohibit field testing and commercialization of Terminator seed technology." In May 2008 worries surfaced that a global ban on terminator technology would be rescinded at a United Nations' summit on genetic diversity, but the issue was not discussed (74). One concern raised about V-GURT plants is that they would cross-pollinate with non-GE plants such as compatible wild relatives or crops in fields of farmers not wishing to adopt GE crops, and become sterile. Although V-GURT plants were sterile, some worried that the sterility trait would occasionally not be expressed, become activated, and cause sterility. Given the complexity of the technology, sterility in non-GE plants caused by cross-pollination would be highly unlikely to occur.

A positive aspect to using such technologies is to inhibit effectively the flow of undesirable GE traits to compatible relatives. It might be prudent, for example, to limit the flow of genes that could give growth or pest-resistance advantages to wild relatives or that encode vaccines, antibodies, or industrial chemicals. Use of V-GURTs thus would slow the movement of GE traits, which could be particularly important in regions of high genetic diversity.

3.3. Why Are Genetically Engineered Crops Patented? Does This Affect Farmers in the United States or Developing Countries?

Companies developing the new GE crops invest substantial amounts of time and money in the research, development, and regulatory approvals (Section 3.5) needed to bring products to market. IP rights provide legal protection for ideas and products (180) and these rights have been key to securing the economic returns necessary to compensate for the substantial investments required to market GE crops (14). Patents can also ensure that results and techniques needed for inventions are ultimately made public, although actual use of the IP is restricted to license holders of the technology. Without this protection, situations could arise where findings are not published and processes are kept secret. The legal system provides ways to protect IP through patenting of not only GE crops, but also the tools (e.g., genes, methods) used to create them.

To ensure that investments made in creating these crops are recouped, seed producers require purchasers of patented GE seeds to sign agreements stating that they will not reuse or sell the seed, and thus growers must repurchase seed each year. This situation is not the first instance of farmers' not being able to reuse seed. In the United States in the 1920s the introduction of hybrid maize seed (275) meant that farmers had to buy seeds each year to capture yield benefits (Section 3.2). Although legal agreements were not involved, the hybrid was a type of "biological patent" that prevented replanting because farmers were unable to create hybrid seeds without the inbred parents, which were protected by the companies that produced the hybrid seed.

Despite potential benefits, patenting in many cases has impeded the use of technologies and development of commercial products. It has often been difficult or impossible to obtain the multiple rights needed to develop and market GE crops. One widely publicized example is Golden Rice, where a large number of IP issues had to be resolved before the engineered genes responsible for provitamin A production could be introduced into local varieties in developing countries (180; Part 1, Section 3.21). However, it should be noted that the major problems actually related to material transfer agreements rather than patents because very few relevant patents had been issued in major rice-consuming countries (44).

In the private sector, obtaining enabling rights has often been accomplished by bringing key technologies and materials under the control of the company through mergers and acquisitions (330). Large agricultural biotechnology companies amassed IP assets through these means or through their own research efforts into the development of new GE crops (151). However, the development of GE agricultural products for farmers in developing economies and of GE seed for low acreage crops in developed countries will most likely be performed, if at all, by nonprofit organizations with public funding (19).

The patenting of living organisms was first realized in 1980 when the Supreme Court decided in *Diamond v. Chakrabarty* that living, man-made microorganisms could be patented. In the same year, the Bayh-Dole Act enacted by Congress encouraged U.S. universities to patent innovations and license them to the private sector (213). These decisions led to striking increases in public sector patents and licensing of patents to the private sector. Licensing of inventions to the private sector often prevents public institution researchers from using materials and methods invented within their own walls to further innovate and create improved commercial agricultural materials (73).

No single public institution has the complete set of IP rights needed to ensure the freedom to operate (FTO) to develop a GE product (85). Although problems with regulatory costs (Section 3.5) and public acceptance also exist, FTO is a major barrier to having all the necessary tools to commercialize a GE product. In a study aimed at better understanding this problem, it was revealed that approximately one quarter of patented agricultural biotechnology inventions were actually created in public sector institutions, which is substantially larger than the IP portfolio of any individual agricultural biotechnology company (138). However, that IP is often scattered among institutions and is often licensed exclusively to entities that restrict its use. In general, public sector scientists have patents on most of the technologies needed to develop GE plant varieties; however, in the past these technologies and other materials necessary to create GE varieties have not been protected properly for public sector use. Open publication or careful reservation of patent rights for public and nonprofit use can address this deficiency. Public sector institutions need to systematically retain rights to inventions that can be used for subsistence and specialty crop development and make them available to others for such purposes (19).

To this end, a number of public sector institutions established PIPRA, the Public Intellectual Property Resource for Agriculture. PIPRA developed a public IP assets database, established best practices to guide development of research innovations, and created specific, pooled public sector IP technology packages to facilitate humanitarian and special use objectives. This effort encourages collaborative research efforts among agricultural scientists at different institutions while recognizing the need to protect and share key IP to make contributions to research for the public good. By 2009 PIPRA has brought together IP from more than 40 universities, public agencies, and not-for-profit institutes (238).

FTO: freedom to operate

PIPRA: Public Intellectual Property Resource for Agriculture

3.4. Does the Export Market Affect Decisions by Farmers to Grow Genetically Engineered Crops?

Strict rules regarding GE presence in seeds and foods for international markets are a key driver for the need to segregate crops, but a lack of standardized, internationally accepted marketing standards, testing methods, and protocols poses significant challenges to the smooth operation of the domestic and international agricultural marketplace (233). By the same token, this situation provides a marketing opportunity for those who can successfully navigate the tangle of regulations and deliver acceptable products. The United States must work toward internationally accepted, science-based standards for trade in GE products that include sampling and testing methods and tolerance levels that result in fair trade practices and that lead to unrestricted shipment of products in international markets.

WTO: World Trade Organization

The United States exported ~\$26.8 billion in agricultural products in 2007 (308). The European Union and the United States are each other's major trading partners, resulting in the largest bilateral trade relationship in the world, with combined economies accounting for 37% of world trade (105). Presently the European Union has approved only a few GE maize varieties for cultivation in member states (Section 3.1). Imported foods, however, can contain GE ingredients that have been approved for food and feed and can be labeled non-GMO in the European Union if they contain less than 0.9% GE content; for Japan, the tolerance is <5% and the tolerance is <1% for Australia and New Zealand (304–307). In the European Union there is zero tolerance for food imports containing unapproved GE ingredients. To aid U.S. farmers in choosing corn hybrids that are acceptable for international trade, the National Corn Growers Association has a database of all GE varieties approved in the United States, indicating their approval status for import into Japan as either food or feed or import into the European Union (219). Despite restrictions on export of GE varieties to certain countries, corn, soybean, and cotton are all grown in the United States for export. A 2006 study reported that 30% of global soybean production is exported and most exports come from countries growing GE soybeans (table 26 in Reference 56). On the basis of size estimates of non-GE soybean markets in the largest non-GE markets of the European Union and Southeast Asia, 10% of global trade in soybeans is estimated to satisfy that market.

The reluctance to import GE crops and products into the European Union relates to a moratorium enacted in late 1998 to prevent U.S. GE corn, cotton, and soybean products from entering European Union markets (327). This moratorium led the U.S. government to file a formal complaint with the World Trade Organization (WTO). The WTO ruled that 24 of the 27 approval procedures for GE crops imposed undue delays in the European Union (327). Europe now faces increased pressure to allow planting of GE crops. The European Union did not appeal the WTO's decision that the de facto moratorium and product-specific approval policies were inconsistent with "sufficient scientific evidence" and "risk assessment requirements" dictated by the SPS (Sanitary and Phytosanitary) Agreement (328).

In a 2008 background paper, the German food and feed industry associations expressed industries' concerns about the negative implications of the European Union biotechnology policy of zero tolerance for GE varieties that are not yet approved in the European Union (1). The stated industry view was that "adhering to a zero-tolerance rule is not possible in international trade with agricultural commodities" and further that "the German and European food and feed industry will no longer be in the position to obtain input materials on the world market." They called for establishing a tolerance for marginal content of GE varieties not yet approved in the European Union, a stance also supported by the European Union Agricultural Commissioner. The need to establish tolerances might be driven by rising food and feed prices and also the possible needs for bioenergy production (292).

What is the current situation with regard to exports? In 2006 the United States, which is the largest producer of corn, provided 42% of the world's supply of maize; of the 56 million metric tons produced, ~20%, was exported (293). Thirty million metric tons of soybean and 260 thousand metric tons of cottonmeal were exported (301). Agricultural exports in 2009 will be worth an estimated \$113 billion, with corn representing \$12.8 billion (302). In fact, the leading exporters of corn, i.e., United States, Argentina, South Africa, and Canada, responsible for 80% of the trade, are all growing GE corn (56). There is a limited non-GE market for corn, mainly in the European Union and to a lesser extent Japan and South Korea, which necessitates segregation of exports for these countries. Approximately 26% of global cotton production was traded, but there appears little effort to segregate GE and non-GE cotton (56). In agreement with these analyses, another 2006 study concluded that export markets for identity-preserved non-GE crop varieties are fairly small worldwide (134). Also, price differences at the farm gate for non-GE corn, soybean, cotton, and canola are not common and are in general not enough to compensate farmers for growing non-GE varieties. Despite what appears to be a relatively minor impact on exports, the potential for such impacts was given as a reason to rescind deregulation of Roundup Ready® alfalfa (Section 2.13), likely because 75% of exported U.S. alfalfa goes to Japan, which banned GE alfalfa (169).

Japan is the largest importer of corn, 16.5 M tons per year for food and animal feed, most from the United States (293). In 2000 legislation was introduced in Japan to prevent imports of food products that contain GE varieties not yet approved in Japan (303). Testing for GE presence is focused on GE products approved for commercialization in other countries, but not in Japan; if found, such products are rejected, destroyed, or diverted to nonfood uses (196). One repercussion of such regulation is that farmers in developing countries resist growing GE crops because of the fear that consumers in high-income importing regions, such as the European Union and Japan, will reject imports from any country that plants GE varieties (231). The segregation and traceability required to assure compliance further discourage such plantings.

In summary, although there has been limited loss of export revenue from some markets that were closed to GE products, the actual revenue loss is far less than what was predicted because of product substitutions that occur in international markets (276). As long as there is another market for the goods produced and they can be adequately segregated, farmers will be able to sell their commodities. Although acceptance of GE products varies among countries, globalization facilitates market substitutions and this minimizes market acceptance issues for new products. GE crops have realized market fluctuations, but the international commodity market is complex and the impact has been minimal compared with the natural volatility in these marketplaces (276).

3.5. Who Is Commercializing Genetically Engineered Crops and What Is the Outcome?

Data regarding food and environmental safety, submitted by the developers of all GE crops, have undergone testing and regulatory scrutiny by federal regulatory agencies (Part I, Section 2.6). Costs of compliance with the biosafety regulations varies significantly depending on the crop, trait, type of regulatory approval, and in which and how many countries developers seek regulatory approval. Using data from interviews with scientists and regulatory personnel, submitted dossiers over a ten year period, and cost data provided by developers of GE crops, compliance costs for Bt maize were estimated at $7.1 to $15.4 million and for HT maize at $6.1 to $14.5 million (170). These costs are in addition to research and development, intellectual property, and technology transfer costs (Section 3.3). Private and public sector developers face uncertainties in compliance costs and outcomes of biosafety regulatory decisions that result in an impact on the product stream submitted for regulatory review. The impact on public sector efforts is more limited because these developers often lack expertise and the physical and financial resources needed to complete the regulatory process.

With the magnitude of these expenses it is perhaps not surprising that an estimated 80% of all GE traits receiving regulatory approval worldwide are owned or co-owned by four major companies and their subsidiaries, Bayer Cropscience (Monheim am Rhein, Germany), Dupont (Wilmington, Delaware, United States), Monsanto (St. Louis, Missouri, United States), and Syngenta (Basel, Switzerland) (170). Despite a considerable trait pipeline from both public and private sector (186), to date most GE crops on the market harbor Bt and/or HT traits. A summation of public sector products created in or for developing countries also shows a sizeable pipeline of innovation, although most products have not reached commercialization (18). Furthermore, high regulatory costs are an impediment to academic and government research institutions and small businesses participating as major players in the commercialization of GE crops (50). This situation has discouraged the development of other GE traits and the introduction of these traits into crops with limited market size (201) and with further application in developing countries where GE crops could have significant impacts. A review of socioeconomic impact assessments of GE crops in developing countries showed that, on average, the impact has been positive although with significant variability across regions, countries, crops, and traits (273). Outcomes have been limited mostly by institutional, not technological, issues. As safe use continues to be demonstrated, further consideration should be given to controlling costs of biosafety regulation and to enabling technology transfer to developing countries.

3.6. Don't We Produce Enough Food to Feed the World Without Genetically Engineered Crops?

Hunger is a complex problem with no simple answer. It can be viewed from different perspectives. For an entire population, hunger is termed food shortage; for the household, it is termed food poverty; and for the individual it is termed food deprivation. Food shortage is one of the causes of food poverty, which in turn is one of the causes of food deprivation (205). Hunger affects millions each year and is only the most visible, and perhaps painful, symptom of the problem of food shortages; more subtle effects result from nutrient deficiencies, which reduce the quality of life and impair human functioning and development. Severe hunger is most dramatic in Africa and Southeast Asia, but is present in more subtle forms in all populations.

Enough food is estimated to be produced worldwide to feed existing populations; a major problem is that poor people cannot access the available food, which is a major socioeconomic issue (see p. 24 in Reference 71). Food distribution is a critical issue. Food needed by the poor in developing countries is either not affordable or cannot move efficiently from where it is produced to where it is needed. A second issue is that the plentiful food in developed countries comes at an environmental cost, regardless of its production method. Third, food choices affect food sufficiency; for example more than 90 to 95% of energy and protein are used or excreted when animals eat plants (see p. 29 in Reference 71). In the United States it takes ~7 kg of grain to produce 1 kg of pork, 5 kg to produce 1 kg of beef, and 2 to 3 kg to produce 1 kg of eggs or poultry. Fourth, population expansion, at the root of food in sufficiency, is predicted to continue for the foreseeable future, particularly in developing countries (294). A recent United Nations study stated that world population will increase from the current 6.5 billion people to 9.1 billion in 2050. Population in more developed regions will remain at ~1.2 billion. In less-developed regions the 5.3 billion will swell to 7.8 billion; even in 2007 food was not plentiful for the 923 million chronically undernourished in these regions (114), but this problem will worsen in coming decades.

Higher food prices exacerbate food sufficiency problems and result from many causes, e.g., larger global demand for food, local weather-related production problems, increased transportation and on-farm costs, and increased use of some food commodities for bioenergy production. Through improvements in agricultural practices and crops, there has been a steady rise in yields over the past decades, but that rate is beginning to decline (57). New methods and crop species are needed to provide higher yields on the same amount of land in an environmentally friendly manner (181). Yields can be improved through GE crops (Section 3.1), particularly in developing countries where disease and pests take a higher toll on production. In addition, more efficient utilization of inputs and lower pesticide usage are necessary to meet increasing food needs while respecting resources and the environment. Sensible use of GE crops, along with other sustainable agricultural practices (Section 2.17), can help achieve this goal.

Mere production of more food and its equitable distribution, however, is not sufficient. Many poor will still be malnourished. Globally, the three deficiencies that can lead to serious health problems are in vitamin A, iron, and iodine (296). Using GE approaches to create crops with higher levels of minerals and vitamins (135, 269, 334) could, along with other approaches, make progress toward alleviating malnutrition and allow people to lead more productive lives.

Food costs in developed nations are inexpensive. In the United States in 2007 less than 10% of disposable income was spent on food (301a). This situation has lulled consumers in some countries into a sense of security about the easy availability of cheap food, leading them to become complacent about the need to invest in agricultural research. Now that food prices are rising, arguably in part because of the use of feedstocks for bioenergy production (288), people are looking for ways to cut food costs and the focus on global warming is causing them to think more about the carbon footprint of our food system (322). Food is transported long distances in the United States, i.e., 1640 km (1019 mi) per delivery. But transportation is not the major contributor to the carbon footprint; it is food production that contributes 83% of the average U.S. household's carbon footprint for food consumption. Because different food groups have different impacts on greenhouse gas emissions, food choices may have a greater impact than food miles on carbon footprint and certainly on food sufficiency.

4. CONCLUDING REMARKS

The use of genetic engineering opens the door to improving agricultural crops in ways not previously possible. But with this capacity comes the responsibility to proceed with caution, investigating possible outcomes carefully. Conversely, there is also a responsibility to utilize the technology where it can provide improvements to human health and the environment and make farmers' efforts more productive. On the basis of the intensive look at the data in the peer-reviewed literature cited in Parts I and II of this review, it appears that the development of GE crops to date has been responsible and regulatory agencies have, in general, proceeded with caution in releasing GE varieties in the United States.

Although no human activity can be guaranteed 100% safe, the commercial GE crops and products available today are at least as safe as those produced by conventional methods. Particularly with regard to environmental safety, we must stay vigilant in our evaluation of GE crops and their impacts to ensure long-term utility, just as with those created using classical methods. Although we should exercise caution, we should not hold GE crops and products to standards not required for food and feed products produced by other technologies. With the proper balance of caution and scrutiny, we can take advantage of the power of this technology without compromising the health of humans, animals, or the environment.

SUMMARY POINTS

1. Foods consumed today derive from plants and animals in which the genetic makeup has been modified by sexual crosses and mutation. Use of recombinant DNA (rDNA), termed genetic engineering or biotechnology, provides a new tool to make genetic modifications.
2. Technically, researchers can transfer genes using rDNA not only within a species, but also from one kingdom to another. Commercially, only a few crops have been so modified, i.e., canola, corn, cotton, papaya, squash, and soybean; however, many others are in development.

3. The environmental safety of genetically engineered (GE) crops and foods, just as with those created by classical breeding and mutation and grown conventionally or organically, must be evaluated on a case-by-case basis to perform meaningful risk assessments.
4. Information from the peer-reviewed literature on the safety of these products should be considered when growing and consuming foods from these crops. Factors beyond the technical, science-based facts should also be part of the decision-making process.
5. Although scientific testing and governmental regulation can reduce the safety risks of conventionally and organically produced and GE crops and food, 100% safety is not achievable.
6. Robust efforts should be made to conserve and enlarge global genebanks and collections created to preserve precious landraces and wild relatives, which are the foundation for future classical breeding, marker assisted selection, and genetic engineering approaches.
7. On the basis of the bulk of data from field tests and farm surveys, pesticide use for GE crop adopters is lower than for conventional variety users. More importantly, extensive data confirm that the environmental impact is substantially lower.
8. Transfer of transgenes is a larger containment issue than pesticides because genes reproduce in the plant. Generalizations about whether gene flow causes significant environmental or economic risks for conventional, organic or GE crops require case-by-case evaluation.
9. Adequate methods for the coexistence of differing varieties and production methods in agriculture are available and being encouraged worldwide; however, minimum standards, not zero tolerance, for GE presence need to be established for this approach to be attainable.
10. Farmers worldwide have adopted GE crops because of the realized economic benefits (which have been demonstrated in numerous studies), time savings, and ease of agricultural practices. Reluctance to adopt mainly relates to apprehensions about rejection in the export market.

FUTURE ISSUES

1. The introduction of pharmaceutical and industrial proteins into edible genetically engineered crops raises issues that require additional safety and regulatory scrutiny.
2. Coexistence measures that permit farmers to use production methods of their choice while respecting their neighbors' rights to do the same must be practiced to achieve economic viability for all farmers.
3. Interest in and funding for independent, peer-reviewed environmental risk assessments of conventional, organic, and GE foods must be encouraged.
4. Unified, rigorous, fact-based, and economically sustainable governmental regulatory policies must be put in place worldwide to allow public and private sector scientists to participate in creating GE crops.

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The author is not aware of any biases that might be perceived as affecting the objectivity of this review.

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