Why virus resistance in transgenic plants, and are there risks associated with the release of virus-resistant transgenic plants?

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Question: Why engineer resistance to viruses in plants?

Answer: Plant viruses can cause severe damage to crops by substantially reducing vigor, yield, and product quality. Losses of several billion dollars are reported annually in vegetable and fruit crops. Control strategies for virus diseases are often prophylactic and directed essentially against vectors that spread viruses within and between fields. By far, the most effective approach to control viruses relies on the use of resistant cultivars and/or rootstocks. Host-resistance genes have been extensively exploited by traditional breeding techniques for the development of virus-resistant plants. However, host resistance has been identified for a few viruses only and a limited number of commercial elite crop cultivars and rootstocks exhibit useful resistance.

Therefore, engineered resistance is attractive to complement conventional breeding approaches, in particular when resistant material with desired horticultural characteristics has not been developed successfully or when no host resistance sources are known. Actually, the deployment of virus-resistant transgenic plants has become an important strategy to implement effective and sustainable control measures against major virus diseases.

Question: How can plants be engineered for virus resistance?

Answer: Virus resistance is achieved usually through the antiviral pathways of RNA silencing, a natural defense mechanism of plants against viruses. The experimental approach consists of isolating a segment of the viral genome itself and transferring it into the genome of a susceptible plant. Integrating a viral gene fragment into a host genome does not cause disease (the entire viral genome is needed to cause disease). Instead, the plant’s natural antiviral mechanism that acts against a virus by degrading its genetic material in a nucleotide sequence specific manner via a cascade of events involving numerous proteins, including ribonucleases (enzymes that cleave RNA), is activated. This targeted degradation of the genome of an invader virus protects plants from virus infection.

Question: Are virus-resistant transgenic crops released?

Answer: Virus-resistant transgenic squash and papaya cultivars have been developed and commercially released in 1996 and 1998, respectively. These were the first transgenic disease-resistant transgenic crop and first transgenic fruit crop, respectively, to be deregulated and introduced in the market. Squash cultivars are resistant to cucumber mosaic virus, zucchini yellow mosaic virus, and watermelon mosaic virus by expressing the coat-protein gene of these three aphid-borne viruses (Figure 1). Similarly, papaya cultivars expressing the coat-protein gene of papaya ringspot virus are resistant to this aphid-borne virus (Figure 2).

The adoption of virus-resistant transgenic squash cultivars has steadily increased since 1996. In 2005, it was estimated that the national adoption rate was 12 percent with the highest rates in New Jersey (25 percent), Florida (22 percent), Georgia (20 percent), South Carolina (20 percent), and Tennessee (20 percent). The adoption of transgenic papaya cultivars has been very high since the start of their release in Hawaii in 1998 (Figure 3), reaching 60 percent in 2005, essentially because the virus had devastated the local papaya industry.

Question: Are environmental safety issues associated with the constitutive expression of viral genetic elements in transgenic plants?

Answer: The expression of a virus gene fragment in a plant confers resistance to virus infection but also raises environmental safety concerns with regard to the constitutive expression of viral genes, which is known to occur only with certain viruses in a few conventional crops. It is conceivable for a virus infecting a transgenic plant to interact with expression products (RNA and proteins) of the virus gene fragment expressed in that transgenic plant. Such interaction can potentially modify the biological properties of existing viruses or lead to the creation of new virus species with novel characteristics, including increased pathogenicity, expanded host range, and altered vector-mediated transmission specificity.

As a consequence of heteroencapsidation (the encapsidation of a challenge viral genome by coat protein subunits expressed from a viral coat protein gene in a transgenic plant), an otherwise aphid-nontransmissible virus could become transmissible. This phenomenon can occur in transgenic plants expressing detectable levels of viral coat protein but is unlikely to occur in transgenic plants that do not produce detectable levels of viral proteins. Also, a chimeric virus resulting from recombination (the creation of chimeric RNA molecules from distinct RNA segments present in two different molecules, for instance, one in a virus infecting a transgenic plant and one in a viral gene fragment expressed by a transgenic plant) can infect an otherwise nonhost plant.

One has to keep in perspective that heteroencapsidation and recombination are not specific to virus-resistant transgenic plants. These two phenomena are well described in conventional plants subject to mixed virus infection. So, the use of transgenic plants does not present any novel environmental safety issue. Therefore, it is not so much the occurrence but rather the consequences of heteroencapsidation and recombination that should be of prime interest when assessing environmental risks of virus-resistant transgenic plants.

Question: Are there other environmental safety issues associated with virus-resistant transgenic plants?

Answer: Environmental safety issues have also been expressed with regard to the transfer of virus genes from transgenic crops into compatible plant species through pollen flow. It is conceivable that an otherwise susceptible plant could become resistance to virus infection upon successful hybridization and introgression with a transgenic crop expressing a viral gene segment. As a consequence, hybrid progeny could have an increased fitness and eventually become more competitive in their habitat. Gene flow from conventional crops into free-living relatives is well documented for some species. In addition, dispersal of host resistance genes and virus transgenes is identical. Therefore, like for the aforementioned issues, it is not so much the occurrence but rather the consequences of gene flow that should draw attention.

Question: Have safety assessment studies been done and what was learned from them?

Answer: Safety assessment studies have been conducted with virus-resistant transgenic crops over the past 15 years. They addressed the occurrence and consequences of heteroencapsidation, recombination, and gene flow. Such studies with transgenic vegetable and fruit-tree crops provided compelling evidence on the limited significance of heteroencapsidation and recombination, indicating that these two phenomena should be considered negligible with regard to adverse environmental effects. Similarly, gene flow studies with virus-resistant transgenic squash and sugar beet showed limited impact in terms of the potential development of hybrid progeny with an increased competitive advantage in natural habitats. In addition, no plant species, cultivated or free-living, is known to have become an invasive weed pest as a consequence of virus-resistance introgression, whether from a conventional or a transgenic crop.

Question: Will virus-resistant transgenic plants impact human health?

Answer: Effects of virus-resistant transgenic crops on human health relate essentially to allergenicity in terms of the properties of proteins encoded by virus gene segments expressed in transgenic plants. Theoretically, virus-transgene protein products can have stretches of amino acid sequences that are identical to potential epitopes of allergen proteins. As a consequence, transgenic plants expressing virus-gene fragments could conceptually promote new food, contact, or inhalant allergies, or modify the level or nature of intrinsic allergens.

Numerous observations suggest that a viral protein in a transgenic plant does not pose any threat to allergenic safety. Most notable is that virus-infected conventional crops have been consumed by humans with no apparent ill effects since plants and plant products have been food. Also, cross-protection (the deliberate inoculation of plants with mild virus strains to protect plants from economic damage of severe challenge virus strains) has been practiced for many years in some vegetable and fruit crops with no documented adverse effect on human health. In addition, virus-resistant transgenic squash and papaya have been released for a number of years and their products are not known as potential allergens, nor is there evidence of increased allerginicity due to their consumption. Together, allergenicity does not appear to be a significant risk for transgenic crops expressing virus-gene fragments.

Question: Will viruses overcome engineered resistance?

Answer: The stability and durability of the engineered virus resistance are important from a disease management standpoint. It is desirable to release elite cultivars and rootstocks that perform well over time. This is true whether resistance is achieved through conventional breeding or agricultural biotechnology. Thorough screening prior to the release of resistant plants is important to determine the breadth of resistance to virus infection and predict its durability.

Some virus strains are able to overcome engineered resistance because RNA silencing is a defense mechanism based on nucleotide sequence identities. Resistance is more likely to be achieved against virus strains with high sequence homology to the strain(s) from which transgenes are derived than those that are most distantly related. In addition, viruses encode some proteins that can act as suppressors of RNA silencing. This means that viruses can counteract the plant's defense mechanism by interfering with some key steps of the antiviral pathways that lead to resistance. Also, the plant developmental stage and features of virus-derived transgenes, among other factors, can influence the effectiveness of RNA silencing. In other words, activation and regulation of the antiviral pathways of RNA silencing is dynamic. Therefore, monitoring the introduction of virus-resistant transgenic crops and eventual emergence of new viral strains is key to maximize the effectiveness and durability of engineered virus resistance.

Question: What is the bottom line?

Answer: Engineered resistance has expanded the scope of innovative approaches for virus-disease control by providing new tools to develop resistant crop cultivars and rootstocks. This technology has also increased opportunities to implement effective and sustainable disease management strategies, and reduced the use of insecticides for control of insect vectors of viruses. The exploitation of the sequence-specific antiviral pathways of RNA silencing, a potent natural plant defense mechanism against viruses, has facilitated the development of virus-resistant crop plants.

Transgenic squash and papaya are in commercial production. Their adoption rate by growers is increasing since their release over a decade ago. Lessons from the commercial release of virus-resistant squash and papaya, and from field experiments with numerous other transgenic crops engineered for virus resistance, including field safety assessment studies, have conclusively demonstrated that benefits outweigh by far any risks to the environment or human health.

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Figure 1. Summer squash plants infected by cucumber mosaic virus, zucchini yellow mosaic virus, and watermelon mosaic virus. The transgenic plant (upper left) is resistant to these three viruses while the conventional plants are susceptible to single infection by CMV (bottom left), ZYMV (bottom right) and WMV (upper right) (Photo by M. Fuchs).

Figure 2. Papaya plants infected by papaya ringspot virus. The conventional plant (left) is susceptible to virus infection while the transgenic plant (right) is resistant (Photo by J. Ogrodnick).

Figure 3. Orchard of transgenic papaya plants resistant to papaya ringspot virus in Hawaii (Photo by M. Fuchs).