Each crop is very specific in its environmental requirements. To deviate at all will decrease both the desired yield and quality of a product. Added to this, seed or propagation material must possess the genetic characters suited to the environment in which it is grown. Most growing systems will work well horticulturally, but systems can differ considerably in cost. Regardless of the type of system, greenhouse agriculture can be extremely expensive. There is no room for mistakes. The cost of CEA may be more than compensated by the significantly higher productivity of greenhouse agriculture as compared to open field agriculture.

The technology of hydroponic systems is changing rapidly with systems today producing yields never before realized. In the last four years, nearly 40 ha of greenhouses have been built in Colorado, Nevada, and Arizona. Many more hectares are planned, not only in the Southwest, but in Mexico. The future for hydroponics appears more positive today than any time over the last 50 years. I sincerely believe hydroponics will be fashionable again!

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Biotechnology and Horticulture

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Biotechnology exploded onto the scene of crop-based agriculture in 1984 with the first report of inheritance of a foreign gene engineered into plants (Horsch et al., 1984). Prior to this time, many scientists speculated on the use of recombinant DNA techniques discovered in the 1970s to improve organisms, but were limited by the capacity to insert new genetic information into the genome. This initial publication of *Agrobacterium*-mediated transformation, and those that followed shortly thereafter (Horsch et al., 1985) sparked a new industry in plant biotechnology. Clearly, biotechnology is in fashion. Many companies were quick to jump on the "biotech wagon" prompted by claims for the potential to modify crop plants. As is typical for an emerging industry that relies heavily upon new technological innovation, the progress was initially slow. This led to a lack of confidence that biotechnology would have a major impact on production agriculture. On the other hand, when one considers that 10 years elapsed between the first report of a transgenic plant and the first commercial product, the Flavr-Savr® tomato, progress has been nothing short of remarkable. Since that time a number of transgenic plants have been grown commercially (Table 1). To date, all of the major crop species, many of them horticultural, have been successfully transformed. Building on the early reports of *Agrobacterium*-mediated transformation, a variety of novel methods for introducing genes into plants were reported, including the delivery of genes into cells by bombardment with particles coated with DNA (Klein et al., 1987).

By all accounts, 1996 was a critical year in the history of plant biotechnology. For the first time American farmers planted large areas in transgenic plants, including herbicide-tolerant soybeans and insect-resistant cotton. In 1996, more than 2 million acres were planted with cotton engineered to express the *Bt* gene, or about 13% of the total U.S. cotton crop. The *Bt* gene from *Bacillus thuringiensis* encodes a protein that is toxic to cotton bollworm, pink bollworm and tobacco budworm. While there continues to be concern over the potential development of

resistance to the *Bt* gene product, the early results look very promising. The following discussion highlights a few of the recent developments in biotechnology that can be expected to impact the horticulture industry.

Ripening and senescence

The phytohormone ethylene plays a major role in regulating fruit ripening and flower senescence in many important horticultural species. The capacity to manipulate fruit ripening or delay flower senescence through biotechnological approaches is now a reality. Ethylene is synthesized by a well known biochemical pathway from *S*-adenosylmethionine (SAM). First, SAM is converted to 1-aminocyclopropane-1-carboxylic acid (ACC) by the action of ACC synthase. Subsequently, ACC is converted to ethylene by ACC oxidase (Kende, 1993). Both of these enzymes are encoded by large gene families (Liang et al., 1992; Tang et al., 1993) and have been cloned from a number of plant species. The potential for modifying fruit ripening by manipulation of this pathway was first reported by Oeller et al. (1991), who described the expression of an antisense RNA for ACC synthase in tomato, which led to a dramatic reduction in the activity of ACC synthase during fruit development and which in turn led to a delay in fruit ripening that was completely reversible by the application of exogenous ethylene gas. These results clearly indicated that the speed of ripening could be manipulated by genetic engineering. Monsanto approached the question of ethylene control by genetically engineering tomatoes to express a gene from *Pseudomonas* that encodes an enzyme capable of breaking down ACC into metabolites other than ethylene (Klee, 1993), which led to a similar delay in fruit ripening that could also be overcome by application of ethylene.

The senescence of some flowers is also associated with a dramatic increase in the production of ethylene, which plays a role in regulating the processes of senescence (Borochov and Woodson, 1989). The genes encoding ethylene biosynthetic pathway enzymes have been cloned from carnation (Park et al., 1992; Wang and Woodson, 1991) and are expressed concomitantly with the increase in ethylene biosynthesis during senescence (Woodson et al., 1992). Using a cDNA clone

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for ACC oxidase, Savin et al. (1995) produced transgenic carnations expressing antisense ACC oxidase and producing flowers with little detectable ethylene; this extended their vase life by more than 200%. In the future it will be important to regulate the expression of these transgenes so that they are not expressed in all tissues and at all stages of development.

Flower color

As consumers of flowers are attracted to novel products, flower color has been a target for biotechnology. Anthocyanins are the major flower pigment in higher plants and much has been learned concerning the biochemistry and molecular biology of their production. A major target has been the development of blue pigmented flowers in species in which these colors do not naturally occur, such as roses or carnations. Delphinidin is the anthocyanin that normally leads to blue pigmentation. Recently, a group from Florigene in Australia successfully identified and cloned a gene from *Petunia hybrida* encoding a flavonoid 3',5'-hydroxylase, which is required for the biosynthesis of delphinidin (Holton et al., 1993). This provides important information necessary for producing transgenic plants with blue pigmentation in their flowers. However, it is only part of the puzzle. In addition to the anthocyanins present in petal cells, flower color is also a function of intracellular pH. This is under the control of a number of genes, the products of which are not currently known. Therefore, the development of transgenic plants with altered flower color will depend on our ability to understand the biochemistry and molecular biology of intracellular pH control.

Plant architecture

The use of biotechnology to manipulate plant architecture is a real possibility. Recently, a group of scientists have successfully engineered tobacco plants to over express a phytochrome A gene that suppresses the shade avoidance mechanism in plants (Robson et al., 1996). The phytochromes are a family of photoreceptors that function as photoreversible pigments in plants. Under normal conditions, plants respond to the enhanced level of far red light in the shade of other plants by excessive elongation. In commercial greenhouses this leads to rapid elongation of potted plants that frequently requires application of growth retarding chemicals. The manipulation of phytochrome gene expression has tremendous potential to regulate plant stature and reduce the need for chemical growth retardants.

In recent years much has been learned about the genetics of flower architecture. Molecular genetic studies have shown that three classes of homeotic genes control the identity of floral organs (Weigel and Meyerowitz, 1994). Distorted expression of these genes at the wrong place and at the wrong time can lead to a modification in floral organ identity. An example of this is the *Agamous* mutation, which leads to the replacement of stamens with petals to produce a double flower. In the future, cloning homologous genes from important floriculture crops could lead to novel floral architecture and more attractive flowers.

Modification of plant lipids

The genetic engineering of the lipid composition of higher plants could improve nutritional value of crops and the ability of plants to withstand low temperature stress. The identification of desaturases, enzymes that introduce a *cis*-double bond in saturated fatty acids, has led to the production of plants with reduced saturated fatty acids. Ishizaki-Nishizawa et al. (1996) introduced a $\Delta 6$ desaturase from cyanobacteria into tobacco, which led to the introduction of a double bond at the 6-carbon position of linoleic acid to produce linolenic acid. This compound is found in fish oils and is thought to reduce blood cholesterol. Another group from Japan generated transgenic tobacco plants expressing a broad spectrum $\Delta 9$ desaturase gene from cyanobacteria, reducing the level of saturation in fatty acids. This resulted in increased chilling tolerance in tobacco seedlings, suggesting that modification of lipid structure could improve stress tolerance

in plants. The modification of lipids in plants has many other potential applications, including industrial uses such as detergents.

Disease and pest resistance

Plant viruses lead to considerable loss in yields and their control was an early target of biotechnology. A number of approaches have been taken to confer resistance to virus infection. Early work focused on the use of genes derived from the viral genome such as those encoding the coat protein (Fitch and Beachy, 1993). This so-called coat protein-mediated resistance has been very successful and is at the stage of commercial release in crop plants (Table 1). The resistance conferred by expression of coat protein genes is limited to the virus from which the gene originated or closely related viruses. More recently, researchers have developed strategies to confer a broad-spectrum resistance to viruses. Tacke et al. (1996) reported that potatoes engineered to express a gene for the movement protein pr17 of the potato leafroll virus were broadly resistant to a number of unrelated potato viruses. Viral movement proteins are involved in the intercellular spread of multiple types of viruses, and mutated versions of these block systemic infection.

Another unique approach to broad-spectrum virus resistance in plants was recently reported by Ogawa et al. (1996) who described a system in which the mammalian interferon-induced antiviral response was genetically engineered into tobacco plants. This is a two component system involving 2',5'-oligoadenylate synthetase and a 2',5'-oligoadenylate dependent ribonuclease. Double stranded RNAs (viral RNAs) are first activated by the 2',5'-oligoadenylate synthetase and this tags them for degradation by the specific RNase. When tobacco plants were engineered to express both genes, they exhibited complete resistance to cucumber mosaic virus. This is a novel approach in that the system targets the virus for destruction rather than just limiting its spread. This is particularly important, as many horticultural crops are propagated vegetatively.

The development of genetic engineering approaches to insect and disease resistance has also progressed rapidly in recent years. The ability of hydrolytic enzymes to limit fungal growth has been exploited to develop crops that are resistant to several fungal pathogens. The introduction of chitinase genes into plants leads to fungal resistance in several cases (Broglie et al., 1991; Grison et al., 1996; Jach et al., 1992; Lin et al., 1995). There is considerable potential to combine the effects of a number of pathogenesis-related genes in plants harboring more than one foreign gene, producing considerable resistance against a broad-spectrum of fungal pathogens.

Insects account for as much as 15% of the crop loss around the world. Resistance to insects has been a long-standing target for biotechnology approaches, and recent success with the *Bt* gene in cotton indicates a great potential for biotechnological solutions to this problem. In addition to the *Bt* gene, proteinase inhibitor genes have proven to be a reliable mechanism to introduce insect resistance into plants. Hilder et al. (1987) first reported on the potential of these genes to confer insect resistance in tobacco. These results have been extended to other crops including tomato (Johnson et al., 1989) and rice (Duan et al., 1996). Certainly the commercial development of these plants is a real possibility.

Table 1. Genetically engineered crops for release in the United States [modified from Astwood and Fuchs (1996)].

Crop	Trait/Product	Organization	Year of introduction
Tomato	Flavr-Savr®	Calgene, Inc.	1994
Squash	Virus-resistant	Asgrow Seed Co.	1995
Tomato	Delayed-ripening	DNA Plant Technology	1995
Tomato	Delayed-softening	Zeneca Seeds, Inc.	1995
Cotton	Bromoxynil-resistant	Calgene, Inc.	1995
Soybean	Roundup Ready™	Monsanto Co.	1996
Cotton	Bollgard™	Monsanto Co.	1996
Canola	Roundup Ready™	Monsanto Co.	1996
Canola	Modified oils	Calgene, Inc.	1996
Potato	Modified starch	AVEBE	1997
Squash	Virus (II)-resistant	Asgrow Seeds, Inc.	1997
Cantaloupe	Virus-resistant	Asgrow Seeds, Inc.	1997

The future

While there is little doubt biotechnology is fashionable, it is clearly not a passing fad. The early successes in commercial development of herbicide and insect resistance in crops clearly points to a strong future for biotechnology in horticulture and agriculture. Perhaps of even greater potential is the discovery of new genes and the processes they regulate through biotechnology research. Current programs in genome mapping and genome sequencing are leading to significant discoveries that will have a significant impact on agriculture in the next century. This is a wondrous time to be a horticultural scientist.

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