PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

# Commercialization of Genetically Engineered Crops

Robert B. Horsch

*Phil. Trans. R. Soc. Lond. B* 1993 **342**, 287-291
doi: 10.1098/rstb.1993.0159

# Commercialization of genetically engineered crops

ROBERT B. HORSCH

*Monsanto Company, 700 Chesterfield Parkway North, St Louis, Missouri 63198, U.S.A.*

## SUMMARY

More abundant harvests from insect- and disease-resistant crops, vine-ripened tomatoes, or less oily potato chips or french fries are some of the benefits that will result from single gene improvements under development today. These single gene traits will be combined with the best new varieties produced by traditional plant breeding and will accelerate the pace and the scope of our ability to develop even better and more productive crops in the future.

The initial group of genetic improvements were first field tested in 1987, improved upon over the past 6 years, and are finally approaching the first commercial sales over the next 3 to 4 years. The key hurdles from discovery of a promising lead to a commercial product trait include: (i) gene cloning and expression; (ii) product development; (iii) field testing; (iv) breeding into multiple elite varieties; (v) product characterization and regulatory review; (vi) public acceptance; and (vii) marketing.

The expense and risk to bring transformed crops to market successfully is significantly higher than for traditionally developed new varieties. The high value of some single gene targets and the possibility for patent protection of the processes and final products provide the incentive for private investment in this area. The value to farmers, consumers, the environment and society in general is very high because the problems being solved are those that have resisted previous attempts through conventional means. Public investment in basic plant science research and private investment in product development is a powerful combination for continual improvement in lowering the cost and improving the quality of the world's food supply.

## 1. NEW PRODUCTS FROM SINGLE GENES

These genetically engineered improvements are not dramatically different from the types of improvements developed by plant breeders since mankind first began deliberately to plant seeds from one plant instead of another because of some beneficial characteristic. During this century, plant breeders have been particularly effective at cross-breeding genes from wild species into cultivated varieties of related crops. For the most part, these new genes cause a measurable change in one or more properties of the plant, such as disease resistance, stature, stress tolerance, seed quality, nutritional properties, etc. However, in nearly every case the mode of action of the gene, and sometimes even the number of genes involved, is unknown to the breeder. When breeding for new traits, even ones believed to be determined by a single gene, large blocks of genes are usually transferred from donor plant to recipient. This is because genetic recombination is rare and does not occur at sites that are close together. Thus, breeding results in the introduction of large blocks of new genes, only one or a few of which are desired. The rest of the genes are tolerated and usually without observable effect, although examples of unwanted linkages are well known. The breeder is limited by genes which can be found in closely related, sexually compatible species or races of the crop.

Genetic transformation is a more precise means to transfer genes into plants. Small, completely defined blocks of genes are packaged into a vector which is then introduced into the plant chromosomes by one of several different delivery techniques (Gasser & Fraley 1992) . The block of genes may end up smaller than originally designed due to partial transfer, or may be transferred in several copies. These complications can be defined later and discarded if a simple block of genes is desired, because many of the individual transformed plant lines will be of the desired nature.

The first three genes to be field tested in a transgenic crop conferred resistance to a plant virus, to caterpillars, or to a commonly used herbicide, glyphosate. Each gene was discovered by a different route. The entire virus was cloned in the case of tobacco mosaic virus (Abel *et al.* 1986). The coat protein gene was then sub-cloned, spliced to a plant gene promoter and transferred to transgenic tobacco and tomato. These experiments were designed to discover the mechanism of a phenomenon known as cross-protection, where plants previously infected with one strain of a virus are somehow able to exclude a second strain of the same virus. The coat protein was found to be responsible for the cross-protection phenomenon but not to cause any of the symptoms itself. It confers protection at lower concentrations than are found in virus-infected plants (Nelson *et al.* 1988; Sanders *et al.* 1992).

The caterpillar control protein gene was cloned from a bacterium, *Bacillus thuringiensis* (Bt), known to control caterpillars by means of an abundant crystal

*Phil. Trans. R. Soc. Lond.* B (1993) **342**, 287–291
*Printed in Great Britain*

protein produced with the spores. This natural product has been sold commercially in the form of dried bacterial spore plus crystal powder for over 30 years. The gene that produces the protein was cloned in 1981 by expressing random genes from the *Bacillus* in *E. coli* cultures and testing for the Bt protein with antibodies (Schnepf & Whiteley 1981). The *E. coli* culture that produced the Bt protein thus contained the Bt gene. When plant transformation and gene expression technology were developed a few years later, the gene was tested in transgenic plants. Unfortunately, it did not express in transgenic plants at detectable levels. Several types of incompatibility were found to be problematic with the bacterial gene sequence, but these were solvable by shortening the gene and/or changing the DNA base sequence without changing the protein itself (Fischhoff *et al.* 1987; Perlak *et al.* 1990).

The herbicide resistance gene was discovered from a detailed study of the biochemical mode of action of the herbicide glyphosate. A suspension culture of petunia cells was selected for growth in the presence of normally inhibitory concentrations of glyphosate. These adapted cells were found to overproduce the target enzyme, EPSPS. This culture was used to purify enough protein to obtain a partial amino acid sequence. The sequence was used to design a set of degenerate DNA probes that were used to clone the gene. Transgenic petunias that over-expressed the EPSPS enzyme were shown to be tolerant to normally lethal concentrations of glyphosate (Shah *et al.* 1986). Unfortunately, the petunia protein did not confer sufficient tolerance to glyphosate in transgenic plants to be agronomically useful. Consequently the EPSPS proteins from a number of other organisms were surveyed to find ones with better tolerance to the herbicide. The petunia gene was then used as a probe to clone EPSPS genes that conferred greater tolerance (Barry *et al.* 1992).

Many more genes have been discovered, cloned and expressed in transgenic plants over the last decade. A wide range of cloning methods have been used. The technologies for gene discovery and cloning get easier every year. The biggest limitation today is knowing what genes to pursue. Some of these new product leads are discussed in this volume. Other examples include genes for nematode or fungal pest resistance, tolerance to environmental stresses, or improvements in processing or food quality (see Fraley 1992).

## 2. BENEFITS TO FARMERS

These agronomic traits will provide farmers new options for protecting their harvests against losses from pests. The most obvious benefits will be in the form of lower total input costs which can be directly measured against current spending on agrochemicals that will be eliminated when using the improved crop varieties. While the price of seed will go up, the total cost of seed plus agrochemicals will go down, resulting in very real cost savings to farmers. Other measurable savings will accrue from reduced application costs such as fuel, time, and machinery use. Although agrochemicals usually provide excellent protection today if used at maximum rates, it is often not economical to do so, resulting in some losses. The built-in pest tolerance should thus provide a higher average level of pest control, resulting in a higher average yield. The improved crop varieties also are a natural fit into the trend towards integrated pest management practices, and may enable savings to be made in secondary costs.

## 3. BENEFITS TO SOCIETY

The effective gene pool potentially available to plant breeders is now larger through the use of gene cloning, gene splicing, and gene transfer technologies. For the most part, finding and using these genes begins with knowledge about their mode of action or with a screen for specific biological activity. One of the most important sources of beneficial genes for crop improvement is the vast gene pool contained in the many diverse species of plants and other organisms that live and grow around the world today. Our capacity to study, to understand, and to find the most useful new genes from nature is still very limited, but it is growing steadily each year. Even as we are just beginning to tap this huge and valuable resource, it is being destroyed by loss of natural habitat and environmental damage.

The terrible irony is that effective use of the biodiversity on earth is one of the single most powerful means to protect it, if we act before it is lost. The products of biotechnology will provide major gains in agricultural productivity and effective biological means to prevent and to remove pollution. The more productive we can be with good crop land, the less natural habitat (which is usually poor farm land anyway) will need to be cleared and ploughed for farming. Other environmental benefits will accrue as well. A major benefit will be improved weed management systems that reduce the need to disturb the soil by ploughing. Ploughing results in enormous loss of top soil from erosion, reducing the capacity of the land to grow food sustainably and damaging water-ways with silt. Genes for herbicide resistance allow for use of the most effective, lowest cost and most environmentally friendly herbicides that would otherwise damage the crop as well as the weeds.

Built-in resistance to insect pests and viruses will also reduce the need to spray insecticides, avoiding the risk of worker exposure, food residues and ground water contamination. It will lower costs to farmers and save fuel and wear and tear on tractors by reducing the number of trips across the field to apply insecticides. These types of saving in production costs have invariably been passed along to consumers in the form of lower prices, increased abundance and greater availability of diverse types of foods that have better quality and appearance.

## 4. INVESTMENT IN RESEARCH AND DEVELOPMENT

The initial wave of genetic improvements such as insect, virus and herbicide resistance were first field tested in 1987 and should finally reach the market




PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

during the next 3 to 4 years. Some of the key hurdles from discovery of a promising gene to a commercial product are: (i) gene improvement and vector construction; (ii) product development; (iii) field testing; (iv) breeding into multiple elite varieties; (v) product characterization and regulatory review; (vi) public acceptance; and (vii) marketing. Each step is expensive, uncertain and time consuming. The profit made during the product's commercial life must pay for the direct expenses of its development and marketing, cover the costs of the leads that failed at some point, and provide more return than putting the same capital into the bank or other investments.

Both public and private investments have been made in plant breeding programs, resulting in benefits to society far in excess of the original investments. While most of the value derived from these research investments have been essentially public domain from the beginning, there are several types of mechanisms in the United States that have been used to protect new varieties and provide a profit to the investors. The plant variety protection act and plant patents provide for an exclusive period of protection for precisely described plant varieties during which no one can propagate that exact variety. A second mechanism is hybridization where F1 hybrid seed must be purchased from the producer each year because the progeny seed will not breed true and will lose the desirable properties of the hybrid. A third, very recent mechanism that addresses genes and therefore plants containing patented genes, is the United States patent system which provides 17 years of protection for patented genes or processes used during product development. Because many developing countries do not offer patent protection for genes or plant varieties, only hybrid seeds provide a means to protect investments made in developing new varieties. In the absence of hybrids, there is little incentive for private investment in development of new varieties that are locally adapted or even to introduce successful varieties from elsewhere in the hope that they will perform well. In this case, public institutions have provided new varieties, sometimes with remarkable success and sometimes not.

The patent system is an ingenious mechanism to provide for stimulation of private investment and entrepreneurial enterprise while fostering scientific communication and technological progress among competing inventors or institutions. It does this by granting a defined period of exclusive ownership for inventions, but requires public disclosure of how to practice the invention at issuance of the patent. This public disclosure allows all other researchers and inventors in the field to use the invention and associated knowledge to improve on the invention and create inventions of their own. Because the invention is disclosed to the public, once the patent expires, the invention enters the public domain for future generations to use.

## 5. PRODUCT DEVELOPMENT

Insect-resistant cotton is a good example of the time and effort involved. The insect control activity of *Bacillus thuringiensis* strains has been known to be the result of proteins and hence of genes since the early 1960s. The first *Bt* gene was cloned and studied in 1981. The first expression in plants was demonstrated in 1986. It took four generations of gene improvement from 1986 to 1990 to produce a version of the gene that would adequately control the worst caterpillar-type insect pests. It will take 1 to 2 years to complete a field test of large enough scope to make a preliminary selection of a commercial candidate transgenic line. It will take another 4 to 6 years to introgress the best gene into several commercial cultivars of cotton and to increase the volume of seed available before the first commercial sales will be possible. Concurrently, three years of multi-location field testing will be needed to ensure product efficacy under a wide range of environments and geographic regions. Also concurrently, product characterization and regulatory data submission, review and approvals will take between 3 and 5 (or more) years. Then, if everything is still satisfactory, the product can be released for sale and compete against existing products and technology for insect control.

## 6. PUBLIC ACCEPTANCE

Public acceptance is the last major hurdle for the products of the science discussed at this conference. As with any new technology, and especially with products as basic as food, people are naturally cautious about change. The industry formed a working group, the International Food Biotechnology Council, to examine the scientific issues and data needed to assure the safety of foods produced by genetic modification. Their findings were published in December 1990 in 'Regulatory toxicology and pharmacology' (Coulston & Kolbye 1990). This document has served as a foundation for science-based discussions with governmental regulatory agencies around the world. The United States Food and Drug Administration published their preliminary analysis in *Science* (Kessler *et al.* 1992). Issues under consideration, arguments being formulated and debated, and requests for regulatory approvals are published in the Federal Register to allow public comment.

The scientific demonstrations of food safety and review by government agencies will play a critical role in gaining public acceptance for products of this science. However, other factors will be critical as well. Among these are the role that credible experts will play in communicating the issues and results of the tests to the public. Academic and government scientists will be seen as more credible than individuals who work for the companies that will profit from the new products or individuals who work for advocacy groups that raise their money by opposing new technology. Both groups have a direct, vested interest in the controversy.

Clear and understandable consumer information is also a very important part of the acceptance process. This is unfortunately often confused with the issue of labels on food products themselves. Labelling is one of several ways to provide information to consumers, but one which would effectively block many uses of the




PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

technology because the cost of separate storage, transport, processing, distribution and marketing would be higher than the savings that would otherwise accrue. Labels have traditionally been used to educate consumers about nutritional content or ingredients known to be a problem for certain people. As the products under development today do not change the nutritional content or wholesomeness of the food, it should not be necessary to impose a label requirement. Rather, the information can be made available to consumers through brochures or other educational means that still protect the public's right to know.

## 7. TECHNOLOGY TRANSFER TO DEVELOPING COUNTRIES

A number of public and private foundations and institutions have targeted biotechnology as a key technology for solving food production problems in developing countries. The Rockefeller Foundation has funded a major program in Rice Biotechnology. The World Bank, USAID, UNESCO, ORSTOM and others have also launched large funding programs to build institutional capacity for biotechnologies around the world. They have recognized the natural applicability of this technology for use by resource-poor farmers because the product comes packaged in a seed. Persley (1990) provides a good discussion of the fit of biotechnology with international institutions, programs and needs.

Several private companies are also involved in technology transfer projects. One example was begun during 1990, when initial discussions were held between Monsanto Company and USAID about possible ways that private companies could help with technology transfer to developing countries in the agricultural biotechnology field. These discussions led to a proposal which was jointly funded by USAID and Monsanto. The first objective was to find a serious problem that was technically feasible to solve with current technology. The second was to design a program to work towards solving that problem and to transfer the technology at the same time. The third was to find a person and an institution with the capacity to carry the project to completion after the training program.

In many developing countries, decreases in the root and tuber crops due to viral infection can reduce potential production by 20–80% depending on the country. The specific choice of crop and virus was deliberately left open while recruiting a project participant so that the final choice could be made in consultation with the African partner institution. Dr Florence Wambugu from the Kenyan Agricultural Research Institute was selected to head the project and has been working on engineering resistance to sweet potato feathery mottle virus (SPFMV) in sweet potatoes. Dr Wambugu had studied sweet potato viruses in the field throughout east Africa and had become one of the leading experts on this problem (Wambugu 1991). She also had just completed studies on sweet potato regeneration from tissue culture: a key prerequisite to transformation. SPFMV is implicated in many of the most devastating viral disease complexes of sweet potato.

The objectives of this project are to begin the development of virus-resistant sweet potatoes and to transfer the technology to an African institution. The training Dr Wambugu participates in includes: (i) basic training in plant cell and tissue culture as well as in plant molecular biology; (ii) research to improve regeneration technology for sweet potato, with emphasis on African varieties; (iii) development of *Agrobacterium* transformation techniques for sweet potato tissues; (iv) development of selection parameters for kanamycin resistance; (v) coupling the regeneration techniques with transformation; (vi) production of transgenic sweet potatoes with SPFMV vectors. Associated training includes assays to monitor the genes and virus resistance as well as knowledge of the regulatory science and process in the United States.

It will be necessary to develop a program to document the food safety of the improved virus resistant sweet potato before introduction into food uses in Africa. As no food products have yet been marketed in any country, the extent of required testing is not known at this time. While no food safety issues can be foreseen that would hinder the adaptation of this new technology, prudence and public acceptance will certainly dictate a significant degree of product testing. There are a number of international institutions that can provide consultation on food safety issues to KARI and Kenyan government officials.

A recent program launched by USAID at Michigan State University has incorporated programs involving companies such as DNAP (Sondahl 1993) and ICI Seeds (Wilson 1993) with public and private institutions in Indonesia and Costa Rica. A new non-profit foundation, ISAAA has facilitated programs between Monsanto Company and Mexico to develop local potato varieties that are resistant to PVX and PVY, and between Asgrow and Costa Rica to develop virus-resistant melons (Altman & James 1993).

As products derived from biotechnology research approach the market place in developed and developing countries, the issues raised by intellectual property rights, or lack thereof, will become more important. A new paradigm is needed for ensuring the maximum benefits from this new and powerful field of science. In the long run all countries will benefit from clear, enforceable protection for intellectual properties such as improved genes and transgenic crops. This will provide incentive for private investment in the most productive sectors of agriculture by multinational companies and for local entrepenurial companies alike, providing products, food, income and tax revenues for the country. This will free the public institutions to invest more in long range research, orphan crops, education, and extension programs.

The author thanks James Altemus and David Fischhoff for reviewing the manuscript.

## REFERENCES

Abel, P.P., Nelson, R.S., De, B., *et al.* 1986 Delay of disease development in transgenic plants that express the tobacco




PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

mosaic virus coat protein gene *Science, Wash.* **232**, 738–743.

Altman, D.W. & James, C. 1993 Public and private sector partnership through ISAAA for the transfer of plant biotechnology applications. *N. Y. Acad. Sci.* (In the press.)

Barry, G., Kishore G., Padgette S., Taylor, M., *et al.* 1992 Inhibitors of amino acid biosynthesis: strategies for imparting glyphosate tolerance to crop plants. In *Biosynthesis and molecular regulation of amino acids in plants* (ed. B. K. Singh, H. E. Flores & J. C. Shannon), pp. 139145. Am. Soc. of Plant Phys.

Coulston, F. & Kolbye, A.C. Jr (eds) 1990 *Regulatory toxicology and pharmacology.* Official Journal of the International Society of Regulatory Toxicology and Pharmacology, vol. 12, no. 3, part 2.

Fischhoff, D.A., Bowdish, K.S., Perlak, F.J., *et al.* 1987 Insect tolerant transgenic tomato plants. *BioTechnology* **5**, 807–813.

Fraley, R.T. 1992 Sustaining the food supply. *BioTechnology* **10**, 40–43.

Gasser, C.S. & Fraley, R.T. 1992 Transgenic crops. *Scient. Am.* **266**, 62–65.

Kessler, D.A., Taylor, M.R., Maryanski, J.H., Flamm, E.L., Kahl, L.S. 1992 The safety of foods developed by biotechnology. *Science, Wash.* **256**, 1747–1749, 1832.

Nelson, R.S., McCormick, S.M., Delannay, X., *et al.* 1988 Virus tolerance, plant growth, and field performance of transgenic tomato plants expressing coat protein from tobacco mosaic virus. *BioTechnology* **6**, 403–409.

Perlak, F.J., Deaton, R.W., Armstrong, T.A., *et al.* 1990 Insect resistant cotton plants. *BioTechnology* **8**, 939–943.

Persley, G.J. 1990 *Beyond Mendel's garden: biotechnology in the service of world agriculture.* Wallingford: C.A.B. International.

Sanders, P.R., Sammons, B., Kaniewski, *et al.* 1992 Field resistance of transgenic tomatoes expressing the tobacco mosaic virus or tomato mosaic virus coat protein genes. *Phytopathology* **82**, 683–690.

Schnepf, H.E. & Whiteley, H.R. 1981 Cloning and expression of the *Bacillus thuringiensis* crystal protein in *Escherichia coli. Proc. natn. Acad. Sci. U.S.A.* **75**, 2893–2897.

Shah, D.M., Horsch, R.B., Klee, H.J., *et al.* 1986 Engineering herbicide tolerance in transgenic plants. *Science, Wash.* **233**, 478–481.

Sondahl, M. 1993 Newsletter of the Agricultural Biotechnology for Sustainable Productivity Project, Michigan State University, East Lansing. *Biolink* **1** (2).

Wambugu, F. 1991 *In vitro* and epidemiological studies of sweet potato virus diseases in Kenya. Ph.D. thesis, University of Bath.

Wilson, M. 1993 Newsletter of the Agricultural Biotechnology for Sustainable Productivity Project, Michigan State University, East Lansing. *Biolink* **1** (2).





PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES