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TRANSGENIC APPROACHES TO COMBATING PESTS AND DISEASES OF PLANTS

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ABSTRACT

This is a review article on the use of transgenic approaches in combating pests and diseases of plants. This review looked at the guiding regulations of the use transgenic plants; its application in fungal disease control and insect resistance. It also viewed the concern associated with transgenic plants, the advantages and disadvantages of its application and its impact on agricultural plants. Whatever misgivings are associated with transgenic plants the damage sue to diseases infestation and the pest menace along with the ecological and human side effects of chemicals calls for a consideration of this method.

INTRODUCTION

Transgenic is the technique of transferring genetic material from one organism into the DNA of another. In the case of plant, it is the technique of transferring genetic material from one species of plant into another species of plant. Transgenic plants possess a gene or genes that have been transferred from a different species.

The aim is to design plants with specific characteristics by artificial insertion of genes from other species or sometimes entirely different kingdoms.

Varieties containing genes of two distinct plant species are frequently created by classical breeders who deliberately force hybridization between distinct plant species when carrying out interspecific or intergeneric *wide crosses* with the intention of developing disease resistant crop varieties. Classical plant breeders use a number of *in vitro* techniques such as protoplast fusion, embryo rescue or mutagenesis to generate diversity and produce plants that would not exist in nature. Such traditional techniques (used since about 1930) have never been controversial, or been given wide publicity except among professional biologists, and have allowed crop breeders to develop varieties of basic food crop, wheat in particular, which resist devastating plant diseases such as rusts. *Hope* is one such wheat variety bred by E. S. McFadden with a gene from a wild grass. *Hope* saved American wheat growers from devastating stem rust outbreaks in the 1930s.

Methods used in traditional breeding that generate plants with DNA from two species by non-recombinant methods are widely familiar to professional plant scientists, and serve important roles in securing a sustainable future for agriculture by protecting crops from pests and helping land and water to be used more efficiently.(Wikipedia 2009)

Deliberate Creation of Transgenic Plants during Breeding

Production of transgenic plants in wide-crosses by plant breeders has been a vital aspect of conventional plant breeding for about a century. Without it, security of our food supply against losses caused by crop pests and diseases such as rusts and mildews would be severely compromised. The first historically recorded interspecies transgenic cereal hybrid was actually between wheat and rye (Wilson, 1876).

In the 20th century, the introduction of alien germplasm into common foods was repeatedly achieved by traditional crop breeders by artificially overcoming fertility barriers. Novel genetic rearrangements of plant chromosomes, such as insertion of large blocks of rye (Secale) genes into wheat chromosomes ('translocations'), has also been exploited widely for many decades (David, *et al.*1996).

By the late 1930s with the introduction of colchicine, perennial grasses were being hybridized with wheat with the aim of transferring disease resistance and perenniality into annual crops, and large-scale practical use of hybrids was well established, leading on to development of Triticosecale and

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other new transgenic cereal crops. In 1985 Plant Genetic Systems (Ghent, Belgium), founded by Marc Van Montagu and Jeff Schell, was the first company to develop genetically engineered (tobacco) plants with insect tolerance by expressing genes encoding for insecticidal proteins from Bacillus thuringiensis (Bt).(Vaeck, *et al*., 1987)

Genetically Engineered Plants

The intentional creation of transgenic plants by laboratory based recombinant DNA methods is more recent (from the mid-70s on) and has been a controversial development in the field of biotechnology opposed vigorously by many NGOs, and several governments, particularly in the Europe. These transgenic recombinant plants (biotech crops, modern transgenics) are transforming agriculture in those regions that have allowed farmers to adopt them, and the area sown to these crops has continued to grow globally in every years since their first introduction in 1996. As at 2006 there were around 250 million acres of genetically engineered crops being grown commercially in 22 countries. The USA has adopted the technology most widely whereas Europe has almost no genetically engineered crops (Peggy 2008). Plates 1 and 2 are examples of genetically modified crops.

Plate 1 Plums that have been genetically engineered to be resistant to the plum pox virus

Plate 2 Genetically engineered rice

Transgenic plants have been deliberately developed for a variety of reasons: longer shelf life, disease resistance, herbicide resistance, pest resistance, non-biological stress resistances, such as to drought or nitrogen starvation, and nutritional improvement. The first modern recombinant crop approved for sale in the US, in 1994, was the FlavrSavr tomato, which had a longer shelf life.

Genetically modified organisms were prior to the coming of the commercially viable crops as the FlavrSavr tomato, only strictly grown indoors (in laboratories). However, after the introduction of the FlavrSavr tomato, certain GMO-crops as GMO-soy and GMO-corn where in the USA being grown outdoors on large scales.

Commercial factors, especially high regulatory and research costs, have so far restricted modern transgenic crop varieties to major traded commodity crops, but recently R&D projects to enhance crops that are locally important in developing counties are being pursued, such as insect protected cow-pea for Africa and insect protected Brinjal eggplant for India. (Web article, 2009)

Transgenic plants have been used for bioremediation of contaminated soils. Mercury, selenium and organic pollutants such as polychlorinated biphenyls (PCBs) have been removed from soils by transgenic plants containing genes for bacterial enzyme (Meagher, 2000).

Regulation of Transgenic Plants

In the United State, the Coordinated Framework for Regulation of Biotechnology governs the regulation of transgenic organisms, including plants. The three agencies involved are:

USDA Animal and Plant Health Inspection Service - who state that The Biotechnology Regulatory Services (BRS) program of the U.S. Department of Agriculture's (USDA) Animal and Plant Health Inspection Service (APHIS) is responsible for regulating the introduction (importation, interstate movement, and field release) of genetically engineered (GE) organisms that may pose a plant pest risk. BRS exercises this authority through APHIS regulations in Title 7, Code of Federal Regulations, Part 340 under the Plant Protection Act of 2000. APHIS protects agriculture and the environment by ensuring that biotechnology is developed and used in a safe manner. Through a strong regulatory framework, BRS ensures the safe and confined introduction of new GE plants with significant safeguards to prevent the accidental release of any GE material. APHIS has regulated the biotechnology industry since 1987 and has authorized more than 10,000 field tests of GE organisms. In order to emphasize the importance of the program, APHIS established BRS in August 2002 by combining units within the agency that dealt with the regulation of biotechnology. Biotechnology, Federal Regulation, and the U.S. Department of Agriculture, February 2006, USDA-APHIS Fact Sheet

- United States Environmental Protection Agency - evaluates potential environmental impacts, especially for genes which encode for pesticide production
- DHHS, Food and Drug Administration (FDA) - evaluates human health risk if the plant is intended for human consumption

Transgenics for Resistance to Fungal Disease

Much work has been performed to develop transformation techniques for sorghum and the millets (Able *et al*., 2001; Datta *et al*., 1999; Jeoung *et al*., 2002; Kothari *et al*., 2005; O'Kennedy *et al*., 2004, 2006; Zhao *et al*., 2000). The transformation of sorghum for increased resistance to disease has been reviewed by Muthukrishnan *et al*. (2001). Downy mildew (Sclerospora graminicola) Gowda,*et al*. (1995); Oh,*et al*. (1996); Thakur and Mathur (2002). Grain quality and mold resistance Franks (2003); Klein, *et al*. (2001) The approaches that have been taken by researchers using genetic engineering for fungal disease resistance in various crops can be grouped into five general categories (reviewed by Punja, 2001): (1) The expression of pathogenesis-related proteins (PR proteins) that are directly toxic to pathogens or that reduce their growth (Mauch *et al*., 1988). (2) The expression of gene products such as polygalacturonase, oxalic acid, lipase, polyphenols, and phytoalexins that destroy or neutralize a component of the pathogen. (3) The expression of gene products that can potentially enhance the structural defenses in the plant e.g. elevated levels of peroxidase and lignin. (4) The expression of gene products releasing signals that can regulate plant defenses. These include production of specific elicitors, hydrogen peroxide, salicylic acid, and ethylene. Hyper-expression of resistance gene (R) products which are transcriptional factors affecting expression of PR proteins and are involved in the hypersensitive response (killing or necrosis of area infected by the pathogen) has been shown to provide protection against fungal attack. Although the concepts of durable resistance and resistance gene deployment have been current for several decades, durable resistance has remained an elusive goal for most crop improvement programs (Michelmore, 2003).

Transgenics for Insect Resistance

The development of transgenic plants with insect resistance has become very important during the last few years and has been reviewed (Christou *et al*., 2006; Pelegrini and Franco, 2005; Sharma *et al*., 2000, 2004). Researchers addressing host plant resistance against pests and diseases in food crops, both in the field and in storage, face the imposing challenge of enhancing resistance while maintaining the desired nutritional and processing qualities of the grain (Bergvinson and Garcia-Lara, 2004). Work on transformation of sorghum and the development of transgenic plants resistant to insect pests and fungal pathogens has been reviewed by O'Kennedy *et al*. (2006). The transformation of sorghum for increased resistance to pest has been reviewed. Green bug (Schizaphisgraminarum) Nagaraj *et al*., (2005)Midge (Stenodiplosissorghicola) Tao *et al*., 2003 Sorghum shoot fly (Atherigonasoccata) Dhillon, *et al*., 2006, Head smut (Ustilagocrameri) Oh, *et al*,. 1994. Using various transformation techniques, several genes for insect resistance, including the cry gene from Bacillus thuringensis (Bt) and protease inhibitor genes, have been transferred to various crops. The use of plant derived genes for expression in transgenic plants for insect resistance has been reviewed by Babu, *et al*., 2003. Protease inhibitors from plants are of particular interest because they are part of the natural plant defense system against insect attack. Protease inhibitors are also reportedly active against nematode, viral, bacterial, and fungal pathogens; thus, they may have a cumulative protective effect in plants (Haq *et al*., 2004). Genes encoding for inhibitors of alpha- and beta-amylases have been used in a variety of cases to produce insect resistant plants (Ishimoto *et al*., 1996). Various protease inhibitors have been shown to be capable of partially controlling certain weevil species (Girard *et al*., 1998), and may be suitable for conferring resistance in stored grains. Of the protease inhibitors expressed in transgenic plants to date, those involving cysteine proteinase inhibitors have shown the most promising results, probably because most phytophagous insects employ these proteases in their digestive tracts (Haq *et al*., 2004). The use of bifunctional alpha-amylase/trypsin inhibitors in transgenic plants would help minimize the likelihood of the appearance of resistant insect strains. Girijashankar *et al*., 2005 introduced the cry1Ac BT gene driven by a maize wound inducible promoter into sorghum by particle bombardment of shoot apices. The somatic embryos and plants subsequently generated from them produced 1-8 ng delta-endotoxin per gram of fresh leaf tissues and showed partial resistance to attack by the stem borer larvae. The development of transgenic pest resistant varieties with strong insecticidal activities has raised concerns on the development of resistance by insect pests with possible environmental consequences (Babu *et al*., 2003). To overcome development of resistance, the expression of multiple genes in plants for long-term durable resistance to insects has been emphasized (Datta *et al*., 2002). The strategy of using more than one foreign inhibitor in transgenic plants that affect different digestive proteases in the insect seems appropriate (Babu *et al*., 2003). Pyramiding of different genes would reduce the probability of resistance development, since multiple concurrent mutations would be needed in individual insects (Sharma *et al*., 2000, 2004). However, reports on the genetic engineering of pest resistance in sorghum and millets are lacking and this is an area that should be researched, to reduce losses due to insect pests in field and more importantly during grain storage.

Maize and cotton with genetically engineered insect resistance have been extensively cultivated for about ten years now, particularly in the USA. And these sorts of maize varieties are already approved in the European Union as well. The active ingredient used to combat the voracious European corn borer caterpillars actually comes from a bacterium. It is now produced in the maize plants. In early genetically modified varieties it was produced in all plant parts. More recent varieties produce the active ingredients predominantly where it is needed: in the stem. This is where the caterpillars live, beyond the reach of conventional pesticides.

Reports of Bt genes being integrated into plants started emerging as far back as the 1980s, initially with tobacco and tomatoes. Then in 1995 the US authorized the first Bt plant: Bt maize.

The Bt concept was particularly attractive for maize, since it made it possible for the first time to combat the European corn borer caterpillars inside the plant. Bt maize has now been grown on a large scale for over a decade, particularly in the USA. In 2007, insect-resistant Bt maize was grown on 21 per cent of the total maize cultivation area, and Bt maize with a combination of insect and herbicide resistance was grown on a further 28 per cent. Various Bt maize varieties are authorized in the EU as well. In 2007 there was notable cultivation of Bt maize primarily in Spain, where it was grown on around 75,000 hectares.

In recent years there has been increasing focus on another maize pest, the Western corn rootworm that is spreading all the time - including in Europe. Bt maize which is resistant to the Western corn rootworm has been authorised in the USA since 2003 and has been grown on a large scale since then.

Ecological Risks

The potential impact on nearby ecosystems is one of the greatest concerns associated with transgenic plants.

Transgenes have the potential for significant ecological impact if the plants can increase in frequency and persist in natural populations. These concerns are similar to those surrounding conventionally bred plant breeds. Several risk factors should be considered:

- Is the transgenic plant capable of growing outside a cultivated area?
- Can the transgenic plant pass its genes to a local wild species, and are the offspring also fertile?
- Does the introduction of the transgene confer a selective advantage to the plant or to hybrids in the wild?

Many domesticated plants can mate and hybridise with wild relatives when they are grown in proximity, and whatever genes the cultivated plant had can then be passed to the hybrid. This applies equally to transgenic plants and conventionally bred plants, as in either case there are advantageous genes that may have negative consequences to an ecosystem upon release. This is normally not a significant concern, despite fears over 'mutant super weeds' overgrowing local wildlife: although hybrid plants are far from uncommon, in most cases these hybrids are not fertile due to polyploidy, and will not multiply or persist long after the original domestic plant is removed from the environment. However, this does not negate the possibility of a negative impact.

Applications

One of the best-known applications of genetic engineering is the creation of GMOs for food use (genetically modified foods); such foods resist insect pests, bacterial or fungal infection, resist herbicides to improve yield, have longer freshness than otherwise, or have superior nutritional value.

Advantages

The modification of the DNA structures of agricultural crops can increase the growth rates and even resistance to different diseases caused by pathogens and parasites. This is extremely beneficial as it can greatly increase the production of food sources with the usage of fewer resources that would be required to host the world's growing populations. These modified crops would also reduce the usage of chemicals, such as fertilizers and pesticides, and therefore decrease the severity and frequency of the damages produced by these chemical pollution (Wikipedia, 2009). Domesticated animals can undergo the same mechanism. Genetic engineering can also increase the genetic diversity of species populations, especially those that are classified as being endangered. Increase in genetic diversity would enabled these organisms to evolve more efficiently that would allow better adaptation to the ecosystems they inhabit. It would also reduce the vulnerability of certain diseases produced by pathogens, as well as decrease the risk of inbreeding that would produce infertile youths. Genetic engineering can be performed to increase to the efficiency of the ecosystem services provided by the other organisms. For example, the modification of a tree's genes could perhaps increase the root systems of these organisms, reduce the damage produced by flood phenomena through flood mitigation.

Disadvantages

There is also much risks and disadvantages through the process of genetic engineering. Since the modification of the domesticated and farmed organisms are generally stronger or "fitter" than those in the wild, these organisms can devastate other organisms within the ecosystem, if they should escape from human industries. They can easily displace, outcompete, and decrease the population of other organisms through consumption. These organisms can also spread pathogens and parasites to other organisms of the ecosystems that they interact with, or even form parasitic symbiosis relationships themselves. Inbreeding can occur as well, as in some cases the genes of certain organisms have been modified so much that they don't correspond to those of natural species. New chemicals can also be produced by the modified organisms that have capabilities of producing harm to the environment. In the perspectives of humans, genetic engineering can limit the amount of food sources for certain people. Because genes from other organisms are inserted, certain people would not desire to consume the food with the inserted gene due to religion and beliefs. Other unknown risks can also arise that are often feared to be uncontrollable or unpreventable by humans(Wikipedia, 2009).

Agricultural Impact of Transgenic Plants

Out crossing of transgenic plants not only poses potential environmental risks, it may also trouble farmers and food producers. Many countries have different legislations for transgenic and conventional plants as well as the derived food and feed, and consumers demand the freedom of choice to buy GM-derived or conventional products. Therefore, farmers and producers must separate both production chains. This requires coexistence measures on the field level as well as trace ability measures throughout the whole food and feed processing chain. Research projects such as Co-Extra, SIGMEA and Transcontainer investigate how farmers can avoid out crossing and mixing of transgenic and non-transgenic crops, and how processors can ensure and verify the separation of both production chains. (Wikipedia, 2009).

CONCLUSION

In view of the treat to food security by loss of plants to diseases and pests, transgenic approach is a welcome method for reducing this loss.

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