Impact of genetically modified plants on the South African flora

By

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Abstract

The rapid advances of plant biotechnology have led to the production of genetically enhanced plants with altered traits such as resistance to an herbicide or insect. The use of these plants has raised concerns that these crops might pose a risk to agricultural ecosystem. A risk assessment study has therefore been conducted using maize and cotton as examples for South Africa. The aspects that have been considered in this study include a possible transgene transfer from a genetically modified plant carrying a herbicide or insect resistance to wild-type plants, the formation of super-weeds and volunteer-weeds, and the movement of a transgene to cultivated plant species. Information for determining any risk by genetically modified plants was obtained through personal interviews and literature search in libraries and the Internet. As an outcome of the study genetically modified maize can be considered as relatively safe for South Africa whereas cotton with weedy relatives might be more problematic due to the chance of out-crossing, which might require risk limitation strategies reducing a possible gene flow.

SAMEVATTING

Die snelle vooruitgang van plantbiotegnologie het gelei tot die produksie van geneties-verbeterde plante met gewysigde eienskappe soos weerstandbiedendheid teen onkruiddoders en insekte. Die gebruik van geneties gemodifiseerde plante het kommer laat ontstaan dat hierdie gewasse 'n risiko inhou vir die landbou ekosisteem. 'n Risikobepalingstudie is geloods waar mielies en katoen as voorbeelde van Suid-Afrikaanse plante gebruik is. Die aspekte wat in hierdie studie bestudeer is, sluit moontlike transgeniese oordrag vanaf 'n geneties-gewysigde plant wat onkruid- of insekwerende weerstandbiedendheid het na wilde-tipe plante, en die ontstaan van super- en toevallige-onkruide, asook transgeniese oordrag na gekweekte plantspesies. Om die risiko in geneties-gewysigde plante vas te stel is inligting versamel deur persoonlike onderhoude en 'n literatuurstudie te doen in biblioteke en op die Internet. Die navorsingsresultate toon aan dat geneties-gewysigde mielies kan veilig geag word vir gebruik in Suid-Afrika, maar dat katoen en verwante onkruid families problematies mag wees as gevolg van die moontlikheid van uit-kruising. Dit mag die daarstelling van risiko-beperkingstrategieë noodsaak om geen oordrag te voorkom.

Research objectives

Using genetic transformation, researchers have produced transgenic plants with desirable traits such as resistance to an insect or herbicide. Even though it is widely used, many concerns are still being expressed regarding the potential risk associated with genetically modifying crops. The possibility of gene transfer from a genetically enhanced plant expressing a herbicide-resistant gene may, for example, ultimately increase the chance of plant invasion altering interactions in natural communities. It is therefore recommendable to determine the impact of such plants on the environment to ensure the safety of such genetically modified crops.

For this MSc thesis a risk assessment study has been conducted to identify and evaluate the possible risk genetically enhanced maize and cotton plants carrying a herbicide or pest resistance transgene might pose to the South African flora. In particular (1) the possibility of movement of a transgene to a wild population of plants by cross-pollination, (2) the creation of 'super weeds' and (3) the movement of a transgene to cultivated plant species were studied in more detail. The risk assessment study was therefore focused on three primary objectives. These were (1) to collect available data from the literature and Internet about the impact on the flora of genetically modified maize and cotton plants, (2) to evaluate a possible impact of such plants on the South African flora and (3) to provide a possible recommendation for planting of such genetically modified plants in South Africa.

Thesis composition

Chapter 1 of this thesis presents the need for an impact assessment and tries to define the concept 'Impact Assessment'. This chapter also identifies the purpose and outcome of an Impact Assessment and the various definitions of an environmental impact assessment. **Chapter 2** analyses the terminology of biotechnology and genetic modification by transgene insertion and outlines the difference between traditional breeding and genetic modification by plant engineering. It also describes the different techniques used in plant engineering, the current production areas, commercial availability and possible benefits of genetically modified crops. This chapter also explains the different traits of such crops like enhanced herbicide and insect resistance. **Chapter 3** focuses on genetically modified plants and its potential application for Africa. This chapter outlines the current research on genetically modified crops in Africa and also provides information about the benefits of such crops, such as maize and cotton, to developing countries like South Africa. **Chapter 4** focuses on the various risks associated with herbicide and pest-resistant genetically modified crops. This chapter explains the potential risk of spreading a transgene for herbicide resistance to other agricultural crops or weedy relatives. It further outlines the gene flow and the various factors reducing gene flow. This chapter also outlines the possible gene transfer from genetically modified plants to wild species important to South Africa. **Chapter 5** focuses on a survey conducted to collect information about genetically modified plants grown in South Africa including their current use and growth areas. This chapter further describes the results of the survey and the potential risk of genetically modified plants in South Africa, such as out-crossing transferring a resistance gene to cultivated crops and wild relatives of maize and cotton. In **Chapter 6** the **Discussion and conclusion** the possible risk of genetically modified maize and cotton plants for the South African flora is discussed based on the information gained during the study. Furthermore, the possible shortcomings of the South African GMO Act and possible risk limitation strategies are discussed.

Acknowledgements

To God Almighty, whose help and wisdom can never be repaid, I thank You for making the completion of my thesis possible.

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Abbreviations and symbols

Bt	*Bacillus thuringiensis*
DNA	Deoxyribonucleic acid
EIA	Environmental impact assessment
EPA	Environmental protection agency
EPSPS	5-enolpyruvylshikimate-3-phosphate synthase
F1	First filial generation
G	Gossypium
GM	Genetically modified plants
GMO's	Genetically modified organisms
Ha	Hectare
IA	Impact assessment
IRM	Insect resistant management
RR	Round Up Ready
T-DNA	Transfer DNA
Ti	Tumor-inducing
Vir	Virulence
Z	Zea
%	Percentage

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Chapter 1

Impact Assessment

1.1 Need for an impact assessment

There are scientific concerns that genetically modified (GM) crops are a risk for the environment and that the environmental impact of such crops has to be studied in detail before GM crops are produced on a large scale in a specific environment. This information will ultimately allow possible recommendations for the appropriate relatively risk-free growing of GM crops and can help developing countries in particular to preserve and use their resources economically to their best advantage.

This impact assessment study has therefore been focused on identifying and evaluating some of the potential risks these GM crops might have on the South African flora. This includes

- The possibility of gene flow from corn and cotton, which are currently grown still on an experimental scale as commercial GM crops in South Africa, to both their wild and cultivated plant relatives by cross-pollination.
- The possible transfer of herbicide resistance from GM crops to weedy relatives.
- The creation of super weeds by the application of GM crops.
- The possible transfer of insect resistance from GM crops to weedy relatives.

1.2 What is an impact assessment?

An impact assessment helps to support a technology development to stimulate further research in order to remedy any intended and unintended adverse effects of the technology (Anandajayasekaram *et al.*, 1996). The term 'impact assessment' means,

Chapter 1

Impact Assessment

1.1 Need for an impact assessment

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1.2 What is an impact assessment?

An impact assessment helps to support a technology development to stimulate further research in order to remedy any intended and unintended adverse effects of the technology (Anandajayasekaram *et al.*, 1996). The term 'impact assessment' means, however, different things to different people and there is generally no single definition of 'impact assessment'. Porter *et al.* (1980) for example defines impact assessment as the systematic study of the effect on a society, which can also be a plant/animal society that may occur when technology or development projects are introduced, extended or modified. It emphasizes those consequences that are unintended, indirect or delayed. In contrast, Boroush *et al.* (1980) defines impact assessment as a perspective that seeks holistically to inquire into short and long-term effects arising from the interactions of technologies and societal systems. In this study the impact assessment is defined as the assessment of the effect of transgenic plants on the South African flora. It is necessary to determine whether the transgenic maize and cotton have an adverse effect on the South Africa's natural environment and whether any precautionary measures need to be taken.

1.3 Purpose of an impact assessment

The purpose of an impact assessment is to increase the awareness of a possible risk due to the generation of unwanted side effects from technological change. This might have provoked a widespread demand for improved mechanisms to manage or control new technology or development project initiatives (Boroush *et al.*, 1980). Impact assessment will enable policy makers to consider systematically the known options about future technological or project developments. It will further allow them to encourage, discourage, modify, and prepare the institutional infrastructure and to block them when appropriate. Furthermore, impact assessment assumes that the future is not pre-ordained and that it can be shaped in accordance with conscious choices.

The purpose of conducting an impact assessment also depends on when the assessment is done (Anandajayasekaram *et al.*, 1996). An impact assessment can be carried out before initiating any research (*ex-ante*) or after the completion of research activity (*ex-poste*), which would then include technology transfer, which is also applicable for this study. The purpose of undertaking an impact assessment prior to starting a research program is to assist in any planning and priority setting. This will allow studying the likely impact of a proposed research activity project and formulate research priorities by examining the

relative benefits of different research programs. It further assists to identify the optimal combination of research programs. An *ex-ante* assessment can also provide a framework for gathering information to carry out an *ex-poste* evaluation.
Anandajayasekaram *et al.* (1996) identified the following two purposes for conducting an *ex-poste* impact assessment after the completion of the program:

- To provide feedback for researchers and policy-makers.
- To improve the decision making process.

The special appeal of impact assessment is that it is an early warning system based on systematic evaluation conducted ahead of the introduction of the technology or project (Boroush *et al.*, 1980). Even though the most thorough impact assessment cannot possibly anticipate all future impacts and risks of a new technology or project, a comprehensive assessment could, however, narrow the usual vast range of uncertainty by distinguishing what is known from what is unknown.

A well-executed impact assessment could provide the following outcome (Porter *et al.*, 1980):

- Modify the project.
- Stimulate research and technology, particularly to deal with adverse effect of the technology.
- Stimulate research to specify or define risks.
- Identify regulatory and legal changes to promote or control the technology.
- Define intervention experiments to reduce negative or enhance positive consequences.
- Stop the technology.
- Provide a reliable base of information to parties at interest.

1.4 Case studies

Case studies are one of the most useful methods of examining the relationship between research and development of a product and it's associated impacts. Such a case study, using two South African GM crops (maize and cotton) as examples, has been carried out in this thesis. Case studies are generally conducted in conjunction with other methods such as surveys, as done also in this study, and cost-benefit methods (Anandajayasekaram *et al.*, 1996). The primary advantage of case studies is that if carried out in sufficient number and detail, they represent probably the best chance of fully identifying the relationship between research and development activities and the resulting impacts. Case studies are suitable to estimate the impact of past research and development activities and, therefore, they are more suitable for assessing applied research.

1.5 Environmental impact assessment

Environmental impact assessment (EIA) can be defined as an activity designed to identify and predict the impact of an action specifically on the bio-geophysical environment and on man's health and well being and to interpret and communicate information about the impacts (Munn, 1979). This thesis can be considered as an EIA study on transgenic plants mainly maize and cotton. However, there is no general and universally accepted definition of EIA. The following examples illustrate the great diversity of definitions:

- To identify, predict and describe in appropriate terms the pros and cons of a proposed development.
- To assess all relevant environmental and resulting social effects, which would result from a project.

Such definitions provide a broad indication of the objectives of EIA but they illustrate different concepts of EIA.

The main objective of EIA is to provide decision-makers with an account of the implications of the proposed course of action before a decision is made (Clark, 1983). Despite the diversity of techniques, the differences in emphasis, and the varied objectives that characterize impact assessment as practiced in different nations, four important aspects of EIA are increasingly approaching consensus (Erickson, 1994). This includes first seeing the environment as the aggregate of things and conditions that surround or envelop everything including non-living things. Secondly, the value of EIA to be more likely realized in the timely communication of information between individuals conducting the assessment and individuals planning a proposed project. Thirdly, realizing that although many environmental components, processes and attributes are amenable to currently available methods of quantification, many are not. Fourthly, mitigation of significant impacts, which includes the minimization of undesirable impacts and enhancement of desirable impacts and which must be assessed for all possible impacts.

Chapter 2

Genetically Engineered Plants

2.1 Terminology

The term biotechnology has been used broadly to describe all genetic modifications, particularly in the field of agriculture (Shandro, 2000). Genetic engineering is an application of biotechnology involving the transfer of a gene between species in order to encourage the desired trait. Plant genetic research in the last decade has focused on a large number of global agricultural challenges (Ghatnekar and Ghatnekar, 2000). Genetic transformation is the heritable change in a cell or an organism brought about by the insertion of foreign DNA into its genome (Agbios, 2001). With the help of gene technology scientists aim to introduce, enhance or delete particular characteristics of a living thing depending on whether the characteristics are considered desirable or undesirable (CSIRO Australia, 2000).

Plants, in which new pieces of DNA are introduced by means (or methods) other than sexual crossing, are referred to as genetically modified (GM), genetically enhanced or transgenic. The broadest definition of plant genetic engineering is changing the genetic make-up of plants to provide plants, plant products and processes for our needs. In this sense, plant genetic engineering has been around for a very long time (Boulter, 1997). A plant contains transgenes that have been artificially inserted instead of acquiring them through pollination is referred to as a transgenic plant. A transgene refers to a gene or group of foreign genes, from one organism that has been inserted into the genome of a different unrelated organism via biotechnology techniques (agbios, 2001).

Proponents argue that advances in this new technology can produce food with yields to feed the growing world population in the 21st century. Critics are concerned that this technology produces uncertainties about the long-term impact on the environment. A criterion by which the importance of this new technology might be evaluated is its contribution to solving or avoiding deleterious consequences of agricultural production practices. Soil erosion and agricultural residues in soil and water are two such problems, where a solution may in part be made possible with the addition of crop varieties designed to be genetically comparable with broad spectrum, high potency, environmentally safe herbicide (Goodman, 1987). Further, traditional corporations, which are among the main proponents of plant biotechnology, view transgenic crops as a way to reduce dependence on inputs such as pesticides and fertilizers (Altieri, 1999).

2.2 Plant engineering

2.2.1 Genetic engineering vs. traditional breeding

Genetic engineering clearly differs from traditional breeding in a number of ways. Traditional breeding relies primarily on selection, using a natural process of sexual and asexual reproduction. Genetic engineering utilizes a process of insertion of genetic material. This can be via the application of a bacterium as a gene vehicle, a gene gun or other direct gene introduction methods (Hansen, 2000). Traditional breeding has typically only recombined genetic traits within species and between related ones. In contrast, genetic engineering can move fully functional gene traits between completely different sorts of organisms (Regal, 1994). Because of the wide variety of genes available for transfer as transgene and the types of alterations that are possible by molecular techniques, gene technology is inherently different to traditional breeding methods (Rogers and Parks, 1995).

Further, one of the disadvantages of traditional breeding is that this process is extremely time consuming. It can take 10 to 12 years to bring new varieties to the market (Scandizzo, 2002). Also, breeders frequently face a situation in which a resistant gene is closely linked to a gene that adversely affects the quality of a crop that is where the two traits are always inherited together. For example, insect resistance in lettuce plants might be inherited along with a tendency for the lettuce to taste bitter (Designer seeds, 1998). Despite these disadvantages, traditional plant breeding will, however, continue to be a vital tool for improvement of plant crops complementing the strategies of genetic engineering (Keshun, 1999).

2.2.2 Genetic engineering techniques

The process of insertions of foreign genetic material via genetic engineering into the host plant genome and the expression of such material are called plant transformation (Hansen, 2000). This provides plants an opportunity for improving their usefulness (Murray, 1997).

For the last two decades there is much development in plant transformation. There are two main methods of transforming a plant cell or plant tissue. The direct gene transfer is obtained by electroporation, biolistics or micro-injection. The method of electroporation is based on the fact that electric pulses can open the cell membrane and allow penetration of alien DNA. Heat shock, in combination with electroporation, has been resulting in a higher efficiency of the transformation. The advantage of electroporation is that it can be applied to protoplasts in angiosperms. The chemical compound polyethyleneglycol changes the pore size of the cell membrane, which enhances the probability of an alien DNA molecule penetrating into the cell. This method has been used for transfer of DNA to monocots as well as dicots, including transfer of DNA to protoplasts. The biolistic approach is using the principle to shoot particles coated with DNA into selected tissues or cells by a particle gun. The particles may consist of either tungsten or gold carrying the DNA and the size may vary. This method is useful for both monocots and dicots (Simonson and Jorgensen, 1997).

DNA transfer can also be obtained by a biological vehicle such as *Agrobacterium spp.* This technique was the first one and has been used for almost 20 years to produce transgenic plants. Many plants are susceptible to this bacterium (Simonsen and Jorgensen, 1997). Within the last few years this Agrobacterium infection was also amended to allow DNA uptake by cereal tissue. *Agrobacterium tumefaciens* is a naturally occurring pathogenic bacterium in the soil that has the ability to transfer part of its tumor-inducing DNA (T-DNA) located on the *Ti* (tumor-inducing) plasmid into a plant genome, which is required for its survival (Figure 2.1). It can only infect a plant and transfer genes through wounds and causes a characteristic growth called crown gall tumor (Figure 2.2). Researchers have, however, disarmed this tumor-inducing part of DNA and have replaced this DNA with genes coding for useful characteristics (Designer seeds, 1998). During transformation the bacterium binds to a wounded plant cell at specific site. Phenolic compounds exuded by the plant cell activate the virulence genes that control the excision and export of the T-DNA segment from the bacteria to the plant cell. (Galun and Breiman, 1997). The most widely transferred traits so far by this technique are genes coding for resistance to herbicides, pests and pathogens (Dunwell, 1998).

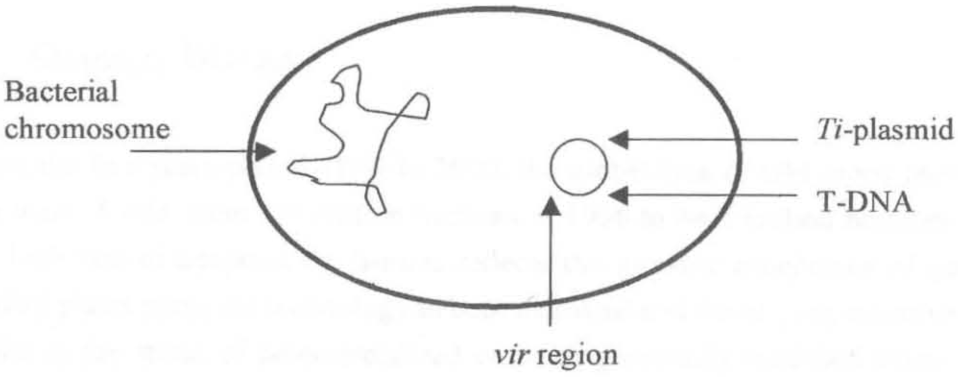

Figure 2.1: *Agrobacterium tumefaciens* cell with *Ti*-plasmid containing T (transfer)-DNA, which is transferred into the plant genome and *vir* (virulence) region, which is required for T-DNA transfer and genome integration.

Figure 2.2: Crown gall tumor on plant. Crown gall disease, which is caused by *Agrobacterium tumefaciens*, produces a tumor-like growth on stems of susceptible plants such as *Kalanchoe* (Source: Designer seeds, 1998).

2.3 Growth of GM crops

During the five years period 1996 to 2000, the global area of GM crops increased by more than 25-fold, from 1.7 million hectares in 1996 to 44.2 million hectares in 2000. This high rate of adoption by farmers reflects the growing acceptance of genetically modified plants using the technology in both industrial and developing countries. Figure 2.3 shows the status of commercialized crops of genetically modified plants in 2000 (James, 2000) and the total acreage has been estimated having increased by 11% when compared to 1999.

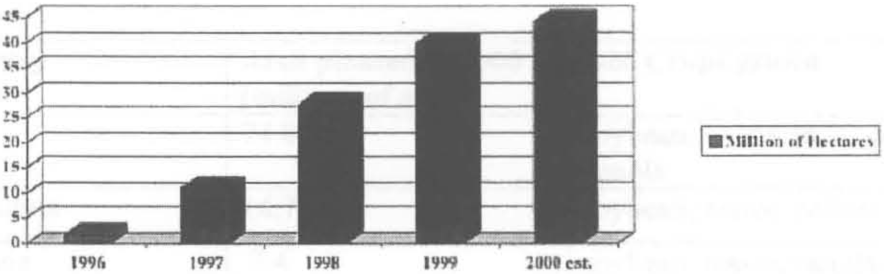

Figure 2.3: The worldwide areas of GM crops over a five-year period. The estimated global area of genetically modified crops for 2000 was 44.2 million hectares.

In 2000, 13 countries grew GM crops (Table 2.1) but four countries had 99% of the global GM crop area. These were the US, Canada, Argentina and China. Among these, the US had the largest GM crop area with 30.3 million hectares followed by Argentina with 10 million hectares, Canada with 3 million and China with 0.5 million hectares. In 2000, herbicide-resistant GM soya occupied 58% (25.8 million hectares) of the global area of GM crops. GM maize followed with 10.3 million hectares, GM cotton covered 5.3 million hectares and GM canola covered 2.8 million hectares. Figure 2.4 shows that out of total worldwide planting, GM soya accounts for 36%, GM cotton for 16%, GM canola for 11% and GM maize accounts for 7% of the total respective crop planting.

Table 2.1: GM crops grown and production area in 2000 (James, 2000).

Country	Area planted in 2000 (millions of acres)	GM Crops grown
USA	74.8	soybean, maize, cotton, canola
Argentina	14.7	soybean, maize, cotton
Canada	7.4	soybean, maize, canola
China	1.2	cotton
South Africa	**0.5**	**maize, cotton**
Australia	0.4	cotton
Mexico	minor	cotton
Bulgaria	minor	maize
Romania	minor	soybean, potato
Spain	minor	maize
Germany	minor	maize
France	minor	maize
Uruguay	minor	soybean

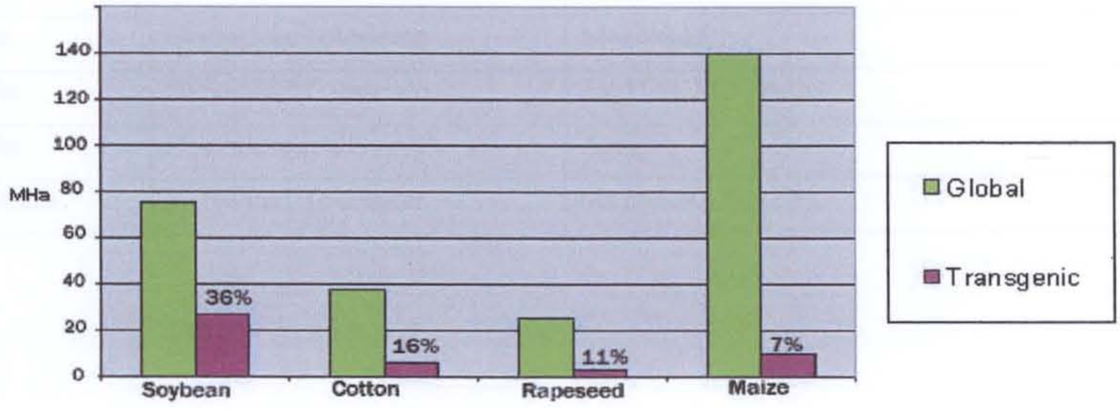

Figure 2.4: Growth of GM crops (Transgenic) in million hectares (M ha) in comparison to total worldwide growth (M ha) of four important crops - soybean, cotton, rapeseed (canola) and maize.

2.4 Traits of GM crops

Plant biotechnology has the capacity to create a greater variety of commercial plants. Genes from sexually incompatible plants or from animals, bacteria or insects can now be introduced into plants. The first transgenic plant, a tobacco plant resistant to an antibiotic, was created in 1983 (BBC News, 1999). Others that followed include Calgene's *Flavr Savr* tomato, a variety of virus- and herbicide resistant crop plants and insect resistant cotton and maize expressing a Bt (*Bacillus thuringiensis*) toxin (Table 2.2). Currently, the major GM crops are soya, canola, cotton and maize (Keshun, 1999).

Table 2.2: Commercially available GM crops.

Crop	Trait	Developed by:
Soya	Herbicide Tolerance	Monsanto, AgrEvo
Soya	Modified Oils	Monsanto, DuPont
Maize	Herbicide Tolerance	Monsanto, AgrEvo
Maize	Hybridization System	AgrEvo
Maize	Insect Resistance	Monsanto, Novartis, AgrEvo
Potato	Insect and Virus Resistance	Monsanto
Cotton	Insect Resistance	Monsanto, AgrEvo, Dow AgroSciences
Cotton	Herbicide Tolerance	Monsanto
Canola	Herbicide Tolerance	AgrEvo, Monsanto
Canola	Hybridization System	AgrEvo
Sugarbeet	Herbicide Tolerance	Monsanto, AgrEvo

Based on the major outcomes of the traits under modification, GM crops fall into two major categories: those with improved agronomic traits and those with altered quality traits (Table 2.3). Herbicide-tolerant crops and Bt-based insect protected crops are best examples of improved agronomic traits (Keshun, 1999). Wilkinson (1997) indicated that about one third of GM crops approved or under review by regulatory agencies for commercialization are with improved agronomic traits including those with tolerance to a broad-spectrum of herbicides. Farmers are usually increasing their acreage of herbicide-resistant crops, because the use of those plants reduces the need to plough, decreases the amount of herbicides and can deliver a cleaner and higher grade of grain and product (Vogt and Parish, 1999).

Table 2.3: Possible benefits of crops with improved agronomic and quality traits.

Crops with improved agronomic traits	**Crops with improved quality traits**
Often reduced use of agrochemicals	Offering new ways to produce raw materials or ingredients
Offering better pest-control tools	Providing healthier food
Achieving high yield potential	Offering superior functionality
Improving crop production efficiency	By-pass processing
Promoting conservation tillage techniques	Facilitate processing
Creating crops which are less harmful to the environment	Offering products with better flavour, colour and taste
Contributing to sustainability of global agriculture	Offering products with longer shelf life
Increasing the world food supply	Improved end-use value

2.4.1 GM crops and herbicide resistance

For an economically superior weed control with higher labour and energy efficiency than the manual or mechanical cultivation methods, herbicides are an indispensable tool of modern agriculture and they are also the targets for GM crops to increase their efficiency. Herbicides are substances that kill plants especially weeds by inducing numerous changes in the growth of plants and their structure. Herbicides reduce manual and mechanical weeding and can also prevent soil erosion. In dry land agriculture, effective herbicidal control ensures higher water availability to the crops and less crop failure due to drought

(Syngenta, 2002). Herbicides may affect the entire plant or only alter particular organs. Presently, different types of herbicides are available. This includes herbicides with either a broad spectrum of action killing crops and weeds indiscriminately (Simonsen and Jorgensen, 1997) or being selective having no or only minor effects on crops. Herbicides might be applied before a crop is planted or when the crop is starting to grow, some herbicides work through the root system of the plants, others through its leaves, some kill the plant on contact, others translocate within the plant.

Various reasons motivated scientists to develop herbicide-resistant GM crops (Le Baron, 1987). This includes:

- Farmers can use the presently available herbicides on additional crops.
- There is a decline in the number of herbicides developed commercially.
- It is time consuming and expensive to develop new herbicides.
- Crops resistant or tolerant to herbicides having long soil residual could be rotated without fear of carry-over injury from the herbicides.
- Weed scientists and plant physiologists have determined the mechanism of action for most herbicides, which is essential before genetic engineering for herbicide selectivity is possible.

The availability of herbicide-resistant GM crops might help farmers to control weeds to a greater extent. But it requires a great deal of administration on the part of farmers (Marshall, 1997). Most common possible risks involved in the introduction of herbicide-resistant GM crops are:

- The potential for over-use of herbicides.
- The risk that herbicide resistance might spread from a transgenic crop to weed species
- The potential for long-term impact on the environment including herbicide residues in soil and water.
- Increased selection for herbicide-resistant weeds by greater reliance on a single herbicide.

- Opportunities for multi-national companies to monopolize the sale of herbicide-resistant GM crop seeds and herbicides through international patents.
- The safety of food harvested from GM crops.

To improve the selectivity of herbicides genes have been identified that express proteins detoxifying herbicides to give crops a selective advantage in the field upon spraying (Stalker, 1989; Andel *et al.*, 1997). Much effort has been put into developing GM crops with tolerance to the non-selective herbicide glyphosate (Figure 2.5). The herbicide glyphosate (phosphonomethyl glycine), which is commonly known as 'Roundup', is one of the most widely used weed control agents today in both developed and developing countries. The herbicide is non-selective and is used in a wide range of applications for total weed control (Ray, 1989). Target of the herbicide is the enzyme EPSPS (5-enolpyruvylshikimate-3-phosphate synthase), which is naturally found in all plants, fungi and bacteria but is absent in animals. The enzyme is an important catalyst in the biochemical pathway for synthesis of the aromatic amino acids phenylalanine, tryptophan and tyrosine (Felsot, 2000). Glyphosate kills plant cells due to the inhibition of the enzyme EPSPS. In GM crops, EPSPS is greatly overproduced due to the insertion of an extra exogenous EPSPS gene into the crop plant allowing the modified plant to survive the herbicide action. However, Glyphosate have no detrimental effect on bacteria and fungi (Carpenter *et al.*, 2002; Busse *et al.*, 2001).

As the metabolic paths of plants differ from animals, glyphosate can be considered as rather safe to humans. Tests have also shown that glyphosate when used according to label directions has no weed killing activity once in contact with the soil. Glyphosate will not move in or on the soil to affect non-target vegetation and it does not move through the soil to enter other non-target plants by the root system. Glyphosate is only effective when it comes into contact with the green growing parts (Annual Report of Monsanto, 2001).

Figure 2.5: Action of the herbicide glyphosate. GM glyphosate-resistant canola surrounded by weeds (left). After spraying of glyphosate only GM canola survives (right; photo courtesy of Monsanto Co.; Source: Virginia Cooperative Extension, 2001).

2.4.2 GM crops and insect resistance

Bt crops are resistant to certain insect pests due to the transfer of a gene to plants from a natural soil bacterium. Bacillus thuringiensis (Bt), which is a common soil bacterium, was first isolated in the Thuringia region of Germany (Cropbiotechnet, 2002). This bacterium has the ability to produce a crystal like protein (delta endotoxin), which can selectively kill Lepidopteran insects. Once the cry protein is eaten, the digestive system of insects activates the toxic form of the protein that can kill the insect within a few days (University of Minnesota, 1997).

Bt toxins are currently the most widely used naturally occurring agricultural pesticide. These organic insecticides are safer and more benign than the chemical pesticide they are replacing and are used extensively both by organic farmers and in transgenic crops to provide safe protection against insect pests (Aroian, 2001). Also due to its target specificity and as it is not synthetic, the organic industry relies heavily on Bt.

Environmental Protection Agency (EPA) in USA classifies Bt pesticides as toxicity III-slightly toxic the same rating as glyphosate (Nelson and Pinto, 2001).

There might be several advantages of using a Bt crop, which includes:

- Improved pest management.
- Reduction in insecticide use.
- Greater yield.
- Improved conditions for non-target organisms.

In 2001, an estimated 12 millions ha of land were planted with crops containing the Bt gene. Table 2.4 shows countries that have commercialized Bt cotton and/or Bt maize.

Table 2.4 Countries that have commercialised Bt cotton and or Bt maize (Source: CropBiotechNet, 2002).

CROP	COUNTRY
Cotton	Argentina Australia China India Indonesia Mexico South Africa United States
Maize	Argentina Canada European Union South Africa United States

Many non-agrochemical organization scientists and farmers around the world are opposed to the use of Bt crops for a variety of reasons including:

- Bt-spray may lose its value as an effective pest management tool if Bt crops are planted extensively. Organic farmers used Bt spray as an effective pesticide control measure for a long time. It has the advantage of Bt toxin breaks down rapidly in the environment and also does not persist in water. Now, because of widespread use of Bt crops in which the Bt does not break down and there is increased exposure to the toxin, insect resistance could develop and this valuable pest management tool could be lost.
- Bt crops may pose serious long-term risks to monarch butterflies.
- Bt crops can contaminate organic and conventional crops and results in loss of bio-diversity
- Bt crops may cause allergic reactions (Pesticide action network update, 2001).

Usage of Bt crops identifies two types of potential risks.

- The development of insect resistance
- The risk to non-target insects (OSTP, 2001)

The lifespan of the Bt product would be significantly shortened if long-term exposure to such insecticide crops were to intensify selection pressure for resistant insects. Insect resistance might also reduce the future utility of naturally occurring microbial Bt. (Levidow *et al.*, 1999).

In order to prevent or mitigate adverse effects, these risk scenarios require:

- Proper monitoring of insect resistance
- Implementation of resistance management plans and
- Examination of potential non-target influence on insects inhabiting the area of Bt maize planting (OSTP, 2001).

There are a variety of insect resistant management (IRM) practices designed to reduce the potential for insect pests to become resistant to a pesticide. The threat that insect resistance presents to the future use of Bt plant pesticide and Bt technology as a whole is a major reason Bt IRM is of great importance. Specific IRM strategies such as the high-dose/ structural refuge strategy will diminish insect resistance to specific Bt proteins produced in maize, cotton and potatoes (Biopesticides registration action document, 2000).

The potential for toxicity to non-target organisms is another area where adverse effects could occur. A Study by Losey *et al.* (1999) shows that Bt-maize pollen can be toxic to monarch larvae when present in significant amounts. Further studies have confirmed that the risk to monarch butterflies from Bt-maize pollen is extremely low (Hansen and Obrycki, 2000).

Chapter 3

GM Crops and Africa

3.1 Plant Biotechnology and GM crops in Africa

Africa missed the green revolution, which helped Asia and Latin America achieve self-sufficiency in food production. The problems of food shortage, a burdened economy, political instability and poor environmental sustainability have contributed to Africa lagging behind in reaping the benefits of the "Green revolution" and it might possibly also miss the "Gene Revolution" (Wambugu, 1999; Woodward *et al.*, 1999)

Biotechnology can make enormous impacts on making intensive agriculture sustainable. Currently, there is misused application of pesticides and poor management of fertilizer over very large areas of intensive agriculture in Africa. With dwindling arable land, the challenge is also to increase yield on current fields and to control pests and diseases that account for about 30% yield losses and to use fertilization more efficiently. Creating plants that carry internal resistance, either through conventional breeding or through application of transgenic approaches to intransigent problems are clearly the most environmentally safe and economically attractive approach. In general, the potential benefits of genetically modified transgenic plants are more crucial to developing nations, whose populations are growing faster than those of developed countries (Keshun, 1999).

There is increasing evidence that genetic modification of plants will contribute to improved agriculture and the quality of life in Africa by producing crops more efficiently and producing healthier food, which is more nutritious, will keep much longer in storage and allows better transport (Flavell, 1999; Bundell, 2000). Africa's crop production per unit area of land is for example the lowest in the world. The average maize yield in Africa is about 1.7 tons per hectare compared to the global average of 4 tons per hectare.

There is a clear potential to already double African crop production by controlling viral diseases, such as maize streak virus, using genetically modified plants (Wambugu, 1999) and improving photosynthesis to increase crop yield (Miflin, 1999). In particular, the use of genetically modified plants might help farmers in Africa to produce more and better food due to new crop varieties that might be drought-tolerant, resistant to insects and weeds and able to use fertilizers more efficiently in Africa with its generally poor soil quality.

One of the most compelling motivations for genetic engineering of plants in Africa is to improve resistance to pest and diseases. The access to novel sources of genetic resistance will provide the opportunity to reduce our dependence on chemical sprays for their control. The use of Bt (*Bacillus thuringiensis*) crop varieties has dramatically reduced the amount of chemical pesticides applied to cotton. In the US, farmers used 450,000 kg less pesticide on Bt cotton than would have been used on conventional varieties in 1998 alone. Further benefits also applicable to Africa include modification of oil, starch and protein to provide sustainable supplies of raw materials for food (Dale, 2000). Recently, sweet potato, a developing country crop, was engineered for improved protein quality (Moffat, 1998). The benefits of genetically engineered plants are shown in Figure 3.1.

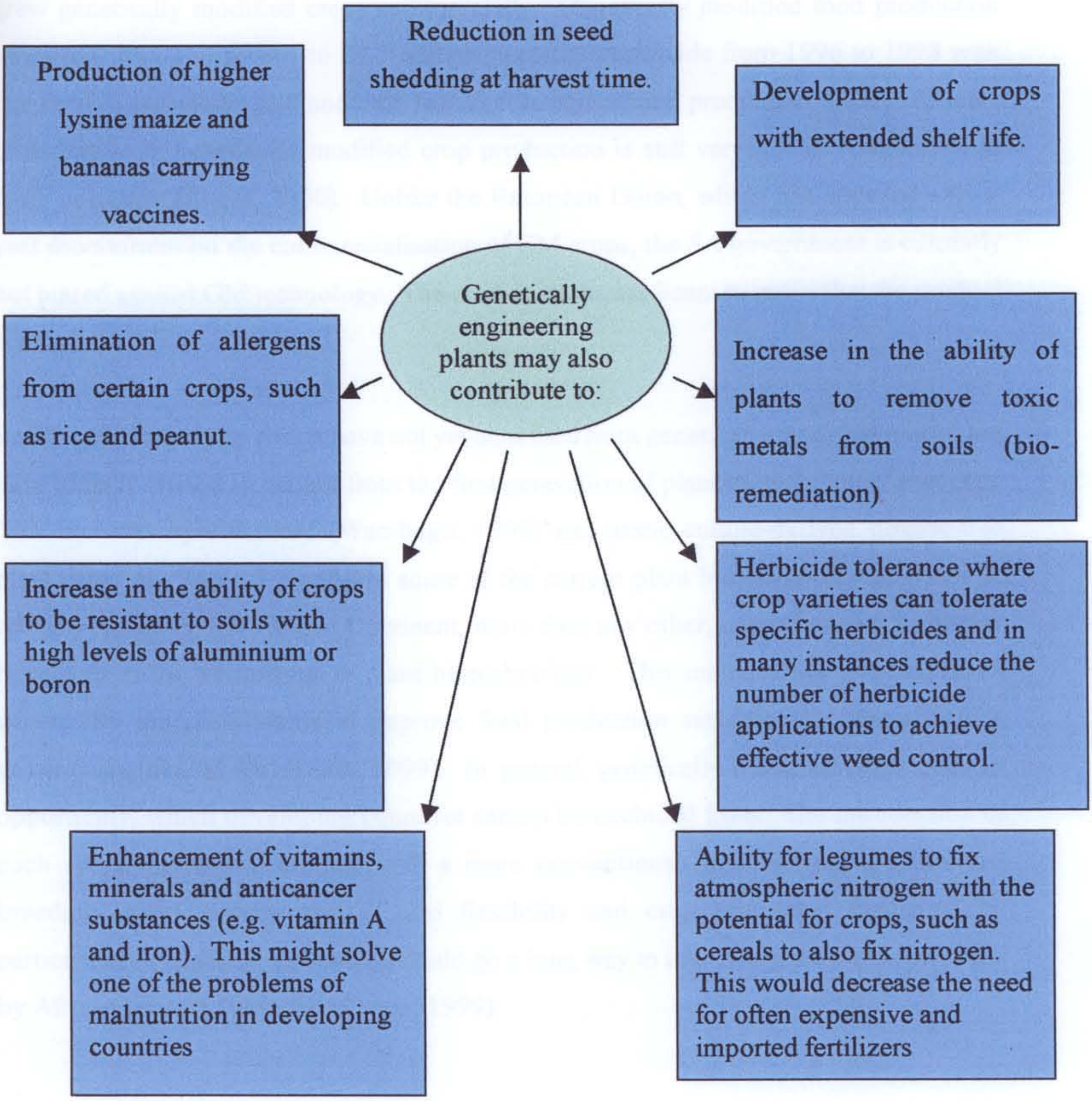

Figure 3.1 Benefits of genetically engineered plant (Skerritt, 2000; Dale, 2000; Feed Magazine, 2000).

In 2000, eight industrialized countries and 5 developing countries including South Africa grew genetically modified crops commercially. Genetically modified food production increased from 1.6 million to 28.3 million hectares worldwide from 1996 to 1998 with measurable economic gain and with sustainable agricultural production. South Africa's contribution in genetically modified crop production is still very low in comparison to other countries (James, 2000). Unlike the European Union, which has imposed a five-year moratorium on the commercialisation of GM crops, the SA government is currently not biased against GM technology. The onus is on the applicant to prove that the product is safe.

Small-scale farmers in Africa have not yet benefited from genetically modified plants, but have already started to benefit from the first generation of plant biotechnology products. This includes hybrid seeds (Wambugu, 1999) and tissue culture-derived disease-free plant material. Table 3.1 outlines some of the current plant biotechnology activities in Africa. However, the African Continent, more than any other, urgently needs to further benefit from the advantages in plant biotechnology. This includes the optimal use of genetically modified plants to improve food production satisfying the demand of a growing population (Anderson, 1999). In general, genetically modified crops offer an opportunity, which developing countries cannot be excluded from. The introduction of such crops, perhaps combined with a more conventional approach using traditional breeding, good management of soil flexibility and crop protection facilitated by participatory extension approaches could go a long way to improving the yields obtained by African farmers (Woodward *et al.*, 1999).

Table 3.1 Current research in plant biotechnology and GM crops in Africa (Brink *et al*., 1998).

Region	**Country**	**Area of research**
North Africa	Egypt	○ Genetic engineering of potatoes, maize and tomatoes
	Morocco	○ Micro propagation of forest trees, date palms ○ Development of disease-free and stress-tolerant plants
	Tunisia	○ Stress-tolerant and disease-resistant plants ○ Micro propagation of date palm, prunes rootstocks and citrus
West Africa	Burkina Faso	○ Biological nitrogen fixation, production of legumes inoculants, medicinal plants
	Cameroon	○ Plant tissue culture ○ Micro propagation of banana
	Gabon	○ Large production of virus-free banana, plantain and cassava plantlets
	Nigeria	○ Micro propagation of cassava, yam and banana ○ DNA fingerprinting of cassava and banana
East Africa	Burundi	○ Micro propagation of ornamental plants-orchids, tissue culture of medicinal plants
	Congo	○ *In vitro* culture of spinach
	Kenya	○ Production of disease-free plants and micro propagation of bananas, potatoes, sweet potato, sugar cane, citrus, papaya
Southern Africa	Malawi	○ Micro propagation of bananas, trees, tropical woody species, tea
	South Africa	○ Genetic engineering of cereals, vegetables and ornamentals, fruits ○ Molecular marker applications
	Zimbabwe	○ Genetic engineering of maize, sorghum and tobacco ○ Micro propagation of cassava, potato and sweet potato ○ Marker-assisted selection
	Zambia	○ Micro propagation of cassava, potato and banana

3.2 GM crops and South Africa

3.2.1 Maize

The South African government gave permission to produce GM crops for human consumption and the first crops were harvested in June/July 2002 (Bissekerm, 2001; Ferreira, 2002). According to GM registrar Shadrack Moephuli, the Bt white maize constitute only 1% of the local market which come from 100 000 ha of farm land scattered around the country. Seed breeder Arthur Schroeder from Pannar believes that GM maize will reach both livestock and people especially the disadvantaged black populations, as maize is their staple food. Genetically engineered yellow Bt maize (Yieldgard) already entered the human food chain in the form of corn flakes and it is often fed to animals (Ferreira, 2002)

3.2.2 Cotton

Cotton cultivation is the main source of income in the Makhathini area in South Africa. The majority of the cotton farmers are smallholders with an average farm size of 1 to 3 ha. Cotton plants account for 90% of the cultivated land. The remaining area is used for cultivating other crops like maize and beans. Insect pests are the main obstacles to increase production. The bollworm complex, namely American bollworm (*Helicoverpa armigera*), Red bollworm (*Diaparopsis castanaea*) and Spiny bollworm (*Erias* spp) are the most damaging insect pests (Ismael *et al.*, 2002).

Genetically modified Bt cotton (Bollgard^{TM}) has been planted as a commercial product since 1997/1998 (Kirsten and Gouse, 2002) and 75 farmers started using the Bt technology with only 80 ha. farms. In 2001/2002, 2976 farmers have decided to use the Bt technology and 5670 ha of land is currently being used for Bt crops (Figure 3.2). This corresponds to about 5% of South Africa's cotton crop (Kirsten and Gouse, 2002). The rate of adoption of Bt crop by small-scale farmers is an indication of the socio-economic benefit of Bt crops in South Africa.

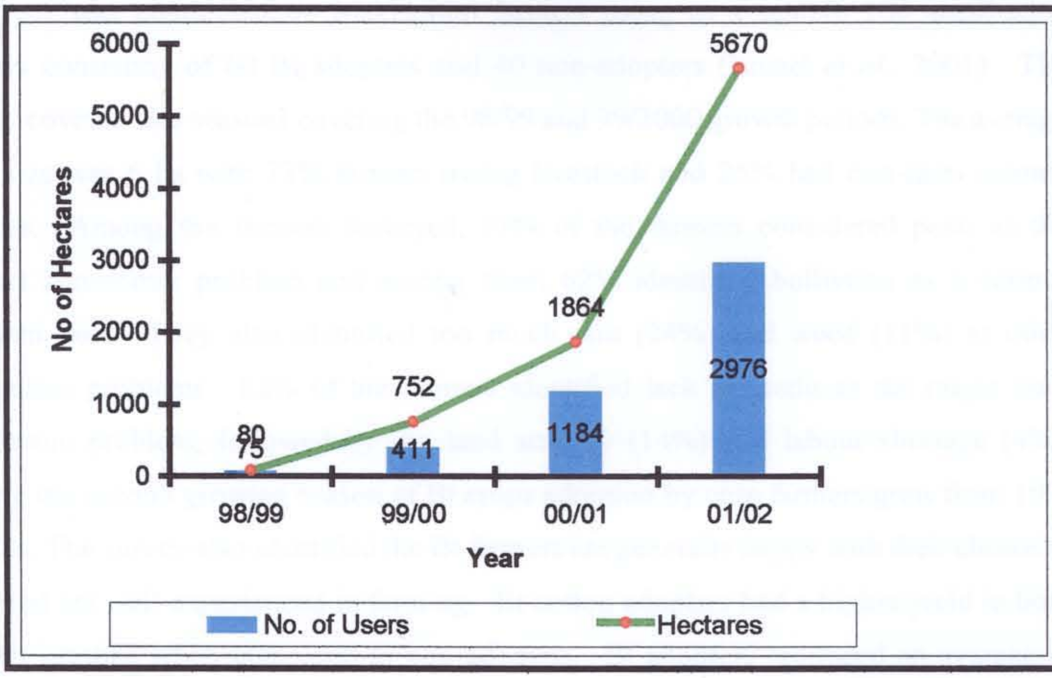

Figure 3.2 Adoption of the Bt technology by small-scale farmers (ISAAA news, 2002).

Makhathini farmers were among the pioneers who tried GM crops in South Africa. They were sceptical when asked not to use chemicals anymore for insect control. Due to the economic hardships, they were willing to try any new technology to make the crop profitable. "Mr. Buthelezi, spokesperson of the Makhathini farmers, testify that the new GM cotton helps them to increase their farm yields substantially and managed to increase the farm to about twice its original size. The GMO Act of 1997 facilitated the commercialisation of GM crops, mainly the insect resistant Bt crops in South Africa. Bt. Cotton is planted in Northern Province, Kwazulu Natal and in the Free State. 1530 commercial farmers and 3000 small-scale farmers planted approximately 100,000 ha in dry land conditions. It is estimated that 95% of the Makhathini farmers will ultimately adopt GM cotton. The main reason for the adoption of cotton is that it is a cash crop and requires less intensive management when compared to other crops in the area, such as beans and maize, and can also survive fluctuating weather (Associated Press Philadelphia, 2001).

A survey was conducted on Makhathini farmers using as a sample 100 small-scale farmers consisting of 60 Bt adopters and 40 non-adopters (Ismael *et al.*, 2001). The survey covered two seasons covering the 98/99 and 99/2000 growth periods. The average farm size was 6 ha with 73% farmers owing livestock and 25% had non-farm income sources. Among the farmers surveyed, 57% of the farmers considered pests as the biggest agronomic problem and among them 62% identified bollworm as a serious problem pest. They also identified too much rain (24%) and weed (11%) as other agronomic problems. 82% of the farmers identified lack of credit as the major non-agronomic problem, followed by the land scarcity (14%) and labour shortage (4%). During the second growing season of Bt crops adoption by crop farmers grew from 19% to 65%. The survey also identified the Bt farmers are generally happy with their choice of crop and are well experienced in farming. Bt cotton adopters had a higher yield in both growth seasons when compared to non-adopters. Bt adopters produced an average of 47kg of cotton per kg of seed while non-adopters produced an average of 30 kg. The average cost of seed for Bt crops was R 206/ha while non-Bt crop farmers spent an average of 123/ha on seeds. At the same period Bt adopters spent an average of R 88/ha on chemical spray compared to the R131/ha by non-Bt farmers. Average gross margin of Bt crop farmers was R 729 compared to R 609 for non-adopters. The survey provided a considerable cause for cautious optimism regarding a higher yield, lower spraying cost, labour and saving time, thus giving hope for optimism regarding the impact of Bt cotton (Associated Press Philadelphia, 2001).

Chapter 4

Risk of GM Crops

4.1 Types of risk

The benefits and risks of genetically engineered plants vary with the specific crop and trait. Even for a specific GM crop, any assessment could vary dramatically from one geographic area to another. Any risk by genetic engineering must be further evaluated in the context of the risk involved in current agricultural practices. Further, rigorous scientific studies are also conducted and reviewed by three agencies of the federal government (Environmental Protection Agency, Food and Drug Administration and Department of Agriculture) before products are commercialized in the US (James and Krattiger, 1996). These testing procedures meet or exceed global standards developed by an international panel of experts formed by the World Health Organization.

4.1.1 Bt crops

Laboratory studies have shown that expression of the Bt gene can affect caterpillars of the monarch butterflies (Losey *et al.*, 1999) when they consume large quantities of Bt corn pollen blown onto milkweeds around maize fields on which the insect is feeding. However, the chance of high amounts of Bt corn pollen settling on milkweeds is rather small (CSIRO Australia, 2000). Another concern is about the development of pest resistance to Bt toxin. The widespread planting of transgenic crops containing the Bt gene will accelerate the development of Bt resistance in pest populations (Gould, 1994). During a study in 1997 in Arizona, scientists projected an increase in resistance to Bt cotton in pink bollworm. In this study, the frequency of a resistant gene in the pink bollworm was about 1 in 10, which was roughly 100-times higher than estimated when compared to other pests of Bt crops. Subsequent studies proved that the estimated frequency of resistance did not increase from 1997 to 1999 (McGinley, 2000). A first strategy to manage insect's resistance includes the planting of 'refuge' or non- Bt crops near Bt cotton, to provide a source of non-resistant target species (in this case *Helicoverpa*) to prevent domination by a non-resistant population. The second strategy is 'pyramiding' which involves creating genetically modified plants with genes of two different toxins. This avoids the risk of loss of single gene resistance (Skerritt, 2000).

A Cornell University study further showed that pollen from Bt maize (altered to contain an insect-killing protein from *Bacillus thuringiensis*) could kill monarch butterfly larvae (Losey *et al.*, 1999). This simple laboratory study would be significant - if monarch larvae eat maize pollen, which they don't. Even dusting of their only food source, milkweed, is unlikely because:

- Larvae may not be present when maize is pollinating.
- Few milkweeds are in and near maize fields.

Bt, which is specific to target pests and harmless to birds, mammals and most other insects, is generally far less risky to monarchs and other beneficial insects than alternative chemical control measures.

A Swiss study also showed that Bt maize could harm beneficial insects including lacewings (Hilbeck *et al.*, 1998a). In this laboratory study, lacewing larvae were fed nothing else but European corn borer larvae, which are killed when they eat Bt maize. For about 21 days, one group of lacewings ate corn borer larvae. However, a large percentage of the lacewings that ate corn borers not subjected to Bt maize also died. They were possibly sick from eating only corn borers, as corn borer larvae are only a minor part of lacewings diet. Interestingly, field studies show that lacewings and other beneficial insects thrive in Bt fields, much better than in fields sprayed with insecticides (Milloy, 1999).

4.1.2 Herbicide-resistant crops

Typical risks for engineered herbicide-resistant crops are shown in Figure 4.1 (Altieri, 1999; Wolfenbarger and Phifer, 2000; Marshall, 1997)

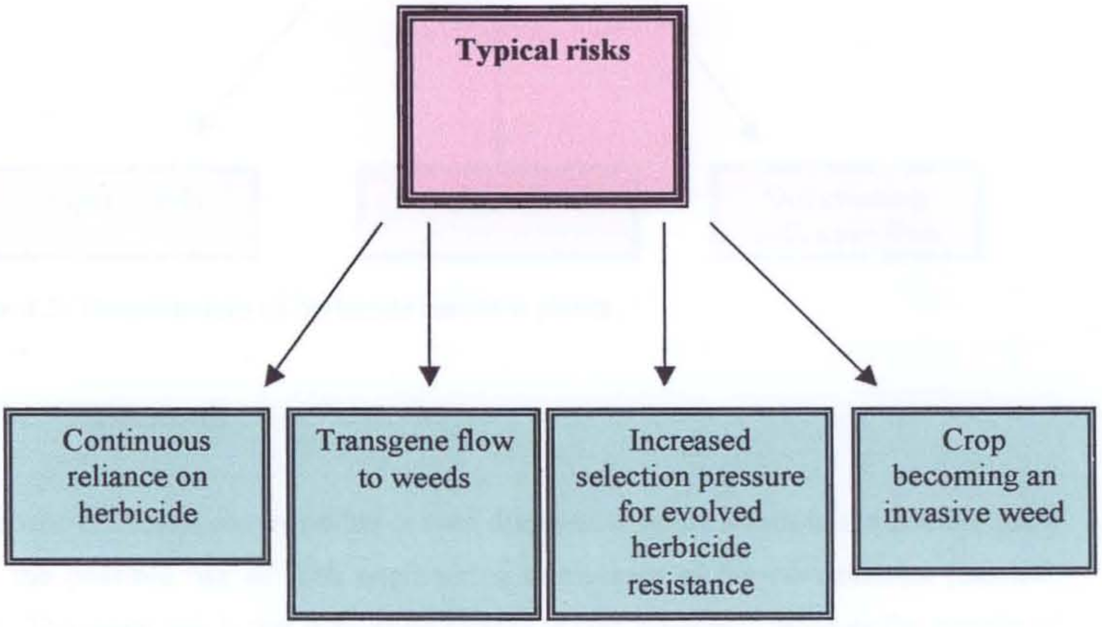

Figure 4.1: Typical risks of herbicide resistant crops. These risks have the potential to offset the prolonged benefits associated with a particular genetically modified crop.

Without doubt, the greatest risk of GM crops is, however, the transgene flow to weedy relatives. The likelihood that a transgene, which increases fitness of a plant, would be accidentally transferred to sexually compatible weeds depends on the degree to which out-crossing can occur in the crop species, and whether there are weedy relatives in the vicinity (FAO conference, 2002). Engineered *Brassica* species, as typical out-crossers, would pose a greater risk than for example engineered soybean, which is almost entirely self-pollinated, or engineered maize, which is grown near a wild relative only in Mexico (Stewart *et al.*, 2002). Possible risks for herbicide-resistant plants are outlined in Figure 4.2.

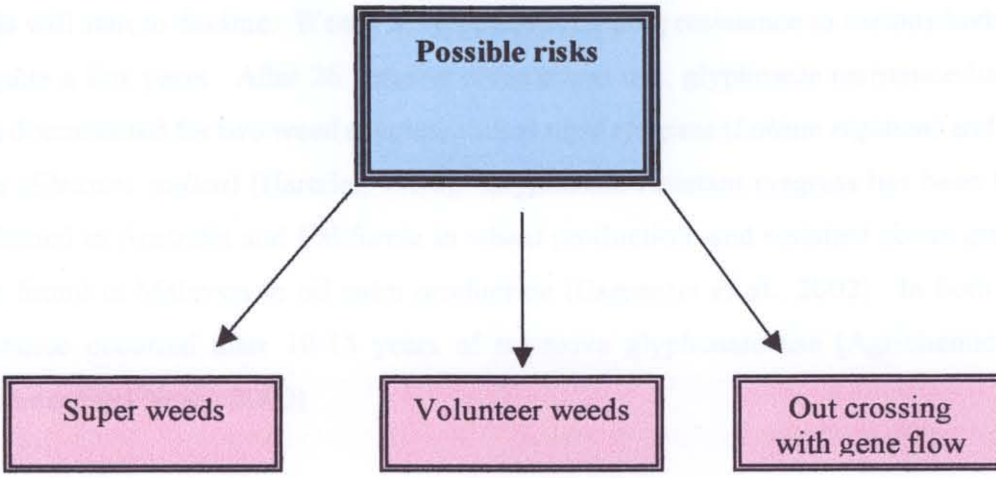

Figure 4.2: Possible risks of herbicide resistant plants.

4.1.2.1 *Super-weeds*

The escape of a transgene by pollen or seed dispersal creating weeds has raised concerns about the possible risk of such engineering technology to the environment (Daniell, 1999). The major risk is that large-scale release of GM crops may promote the transfer of a transgene from crops to wild species by sexual hybridization and the hybrid may become a ''super weed'' (Raybould and Gray, 1994). When a single herbicide is used repeatedly on a crop, the chances of herbicide resistance developing in a weed population greatly increases. The increased herbicide use may lead to reduction in crop yield as two kinds of plant competition occur with the increased use (Altieri, 1999). The genetic engineering of crops that are resistant to herbicides might enable the widespread use of these herbicides without concern for the stage of the crop growing cycle. But these herbicides, although considered to be environmentally safe, would no longer be effective against weeds that had captured a transgene for herbicide resistance, leading to the use of more dangerous chemicals. These herbicide resistant crops could cause the creation of "super-weeds" (Ellstrand and Hoffman, 1990). Other crops would thus be forced to compete with these new super-weeds. For example, populations of annual ryegrass (*Lolium rigidum*) have been found in Australia, which are resistant to herbicides from ten different chemical classes (Rogers and Parkes, 1995). As crops are forced to compete with new super-weeds that no longer succumb to the usual herbicide treatment their yields will start to decline. Weeds have been developing resistance to various herbicides for quite a few years. After 26 years of commercial use, glyphosate resistance has only been documented for two weed species, annual rigid ryegrass (*Lolium rigidum*) and goose grass (*Eleusine indica*) (Hartzler, 1998). Glyphosate-resistant ryegrass has been further confirmed in Australia and California in wheat production, and resistant goose grass has been found in Malaysia in oil palm production (Carpenter *et al.*, 2002). In both cases, resistance occurred after 10-15 years of intensive glyphosate use (Agrichemical and Environmental News, 2000)

It has been found that genetically modified herbicide-tolerant oilseed rape pose a potential risk of spreading herbicide-tolerant genes to other rape crops or to weedy relatives (Squire *et al.*, 1997). Oilseed rape belongs to the *cruciferae* family, is indigenous to Western Europe and is one of the most problematic crops concerning gene flow from the crop to weedy relatives (Kapteijns, 1993). For example, of the 160 species of *Brassicaceae* present in Australia, several species are important weeds of the Southern Australian cropping zone with overlapping flowering time. Among these weeds are *B. rapa, B. juncea, B. tournefortii, diplotaxis tenuifolia* (Lincoln weed) and *Raphanus raphanistrum* (wild radish) (Virtue, 1996). *B. rapus* is derived from the cross between *B. rapa* and *B. oleracea*. *B. juncea*, is a hybrid between *B. rapa* and *B. nigra* and is generally thought to have originated in the Middle East (Reiger *et al.*, 1999). Inadvertent hybridization could generate persistent herbicide-tolerant weeds that would limit the efficiency of the herbicide in rape. Such scenarios could adversely affect overall herbicide usage or could preclude options that are environmentally preferable. Transgene flow can very likely occur in rape, because viable pollen can travel up to 2 km (Timmons *et al.*, 1996). Volunteer rape has, for example, a constant flux with feral rape outside the field, via a flow of pollen and of seeds (Squire *et al.*, 1997). When interspecies hybridization was tested in field experiments studying the initial hybrid and then back crosses with the weed, fertile, weed-like plants were found after just two generations of crosses between *Brassica napus* and *Brassica campestris/rapa* (Jorgensen *et al.*, 1996;

Mikkelsen *et al.*, 1996). When GM oilseed rape with tolerance to three different herbicides was cultivated in close proximity, some progeny had multiple herbicide tolerance (Reboud *et al.*, 1998).

4.1.2.2 *Volunteer-weeds*

The seeds remaining in the soil from the previous season's herbicide-resistant GM crop might germinate the following year in rotational crops. If these "volunteer-weeds" are resistant to herbicides, which are used on the new crop, competition may become a critical yield-limiting factor, because crop yield is dependent on the plants not growing too close together (Rautenberg, 1998). Volunteer crops are already considered to cause significant problems in weed control. Careful considerations must be given to the herbicide/crop combinations sold in a particular country. For example, volunteer potatoes are particularly troublesome in the UK, for which the herbicide glyphosate is a valuable herbicide (Marshall, 1997). However, if glyphosate-tolerant engineered potatoes would be introduced to the UK, their volunteers would undoubtedly become a serious weed problem, given that there are no satisfactory alternative herbicides for their control (Marshall, 1997). The widespread use of herbicide resistant crops is also likely to increase herbicide use, as they have to control the engineered super and volunteer-weeds.

4.1.2.3 *Out-crossing*

4.1.2.3.1 *Gene flow*

As gene flow is a natural process, it is important to improve our understanding of this phenomenon, and modern biotechnology is helping make research into gene flow more accurate and informative. It is the movement of gene mediated by pollen flow and seed dispersal (Rieger *et al.*, 1999) and this is a major process determining the genetic structure of a plant population. Pollen will be the most important vehicle for the escape of transgenes (Rongnli *et al.*, 2000). The difference between gene flow either by pollen or seeds is that seeds usually only transfer cytoplasmic inherited genes (Raamsdonk *et al.*, 1997). The gene transfer is further divided into vertical gene transfer and horizontal gene transfer. Vertical gene transfer is the sexual transfer of genes between two genetic different entities e.g.: two distinct populations of two species. Hybridization action through out-crossing is needed for a successful gene transfer. Out-crossing is thus a prerequisite for gene transfer. Horizontal gene transfer refers to non-sexual gene-transfer among organisms, which may belong to unrelated systematic groups e.g.: gene transfer between higher plants and microbes (Kjellsson *et al.*, 1997). Gene flow within and between populations has an important role in maintaining population genetic structure, enabling adaptation to changing environmental circumstances and reducing vulnerability to evolutionary hazards, such as inbreeding depression and genetic drift (Rieger *et al.*, 1999).

There are various methods used to measure pollen flow. These methods can be classified under two main groups, which are "Indirect" and "Direct" (Real, 1983). Due to the fact that pollen reaches a target from any direction, determining the movements of pollen grains from specific sources is difficult. In the first indirect method, dyes or powders are placed on the anthers and then after a period of pollination activity, surrounding plants are searched for the marker (Real, 1983). Further, indirect methods are secondly where chemicals are introduced into or within the pollen grains. Then after the pollen disperses, surrounding plants are searched for the labeled grains. The movement of all-potential pollinators is observed in a third method so that their behavior and movement patterns can be observed when in contact with pollen (Real, 1983).

As direct methods artificial samplers have been used to understand pollen flight dynamics for forest trees and for wind pollinated crop and weed stands (Ogden *et al.*, 1974). The accuracy and efficiency of these samplers depends on the mechanical design especially on the speed of the air as it passes through the collector and the size and shape of the collecting surface (Ogden *et al.*, 1974). A second method includes the use of stigmas, where counting of pollen grains eliminates the possible sources of error that the mechanical devices create (Real, 1983).

Gene flow is not reserved strictly for GM crops as gene flow has occurred since crop domestication. Gene flow will occur if there are wild relatives in the vicinity of the crop plant (Carpenter *et al.*, 2002). Sorghum and sunflowers have a greater likelihood for cross-pollination with weedy relatives, when these weeds exist where the crop is grown. Should a sorghum or sunflower plant derive through application of genetic engineering, the regulatory review process would require consideration of the potential impact on weediness of wild or weedy relatives (Council for biotechnology information, 2001). The potential for gene flow between weedy species and a related GM crop is a key component of the risk assessment required by regulatory agencies for every new plant variety developed through biotechnology.

In the US and Canada, crops, such as corn and soybeans, do not have wild relatives nearby. Gene flow is therefore limited to neighboring cultivated plants within the same field or to nearby fields. The likelihood of gene flow diminishes the further apart plants are located, even if they are in the same field or region. For some crops, such as soybeans, pollination characteristics limit gene exchange even between neighboring plants in the same field. For others, such as sorghum and sunflowers, more attention must be given to cross-pollination with weedy relatives.

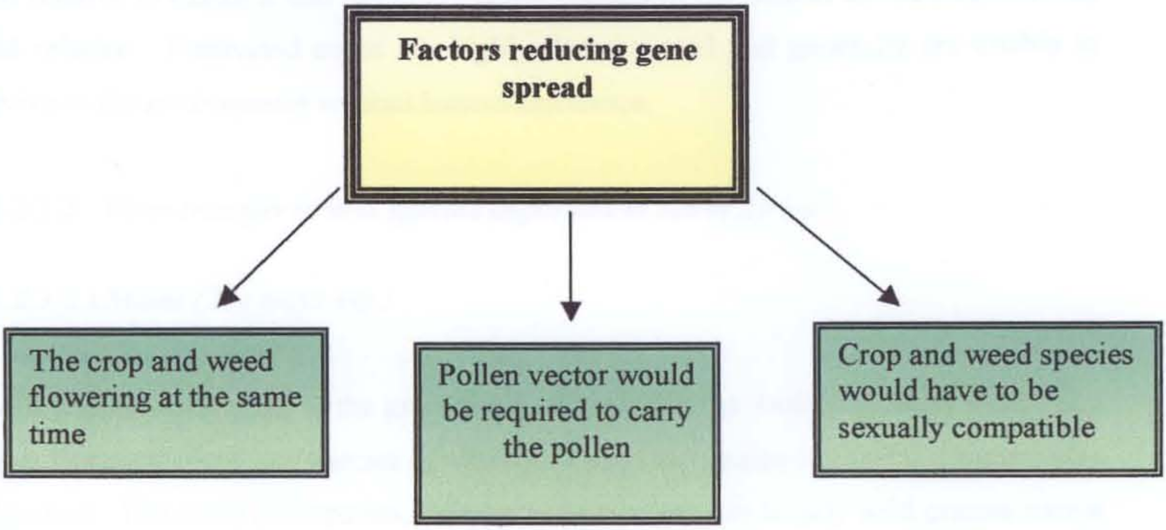

Figure 4.3: Factors reducing gene spread.

The chance of a gene spread is greatly reduced by some factors (Figure 4.3). This includes that (1) the crop and weed would have to flower at the same time, (2) a pollen vector, such as insects or wind, would be required to carry the pollen and (3) the crop and weed species would need to be sexually compatible and capable of producing fertile progeny weed plants. If there is no sexual compatibility between plants, there can be no gene flow - just as a dog cannot successfully mate with a cat. The pollinating characteristics of the particular plant species are important as well.

Some crops, such as maize, are cross-pollinators and can exchange genes relatively easily with other maize plants or with wild relatives under appropriate conditions. Gene flow in self-pollinators, such as wheat and soybeans, occurs infrequently. In addition, the crop must be grown in an area where a wild relative is native. For example, there are no wild relatives of maize or soybean, two of the most widely planted crops, in the US. Gene flow from these crops into wild populations, therefore, does not occur (Rissler and Mellon, 1993). Also, there must be a benefit associated with the gene of interest in order for it to persist. Genetic modifications must increase a plant's ability to survive and reproduce in order for any gene to be actively selected and preserved over generations.

There is little selective advantage to having a trait that confers herbicide-tolerance in a wild relative of maize if that herbicide is not present in the natural environment of that wild relative. Cultivated crops are highly domesticated and generally are unable to survive in the environment without human assistance.

4.1.2.3.2 *Gene transfer to wild species important to South Africa*

4.1.2.3.2.1 *Maize (Zea mays ssp.)*

Zea is a genus belonging to the grass family *Poaceae* in the *Andropogoneae* tribe. The genus *Zea* consists of four species of which *Zea mays* ssp (maize or corn) is economically important. The other *Zea* species, referred to as teosinte, are largely wild grasses native to Mexico and Central America (Doebley, 1990). Teosinte species show little tendency to spread beyond their natural range and distribution is restricted to North, Central and South America. The nearest teosinte relative to *Z. mays* ssp is Z. *mays* ssp. Mexicana Iltis (previously classified as *Euchlaena mexicana*, *Zea mexicana*) (2n = 20) has limited use as a forage and green fodder crop, but can be problematic due to weedy tendencies (Doebley, 1990). This central Mexican annual teosinte is large flowered; mostly weedy with a broad distribution across the central highlands of Mexico.

Gene exchange between cultivated and genetically engineered maize occurs naturally at the present time (Agbios, 2001). Cross-fertilization normally occurs when a transgene-laden pollen is carried by bees or blown with the wind from one field to another. The resulting contamination from GM maize can ruin any normal or organically grown maize crop by rendering traditional hybrid maize worthless for export to countries where consumers are wary of the gene transfer technology (Agbios, 2001).

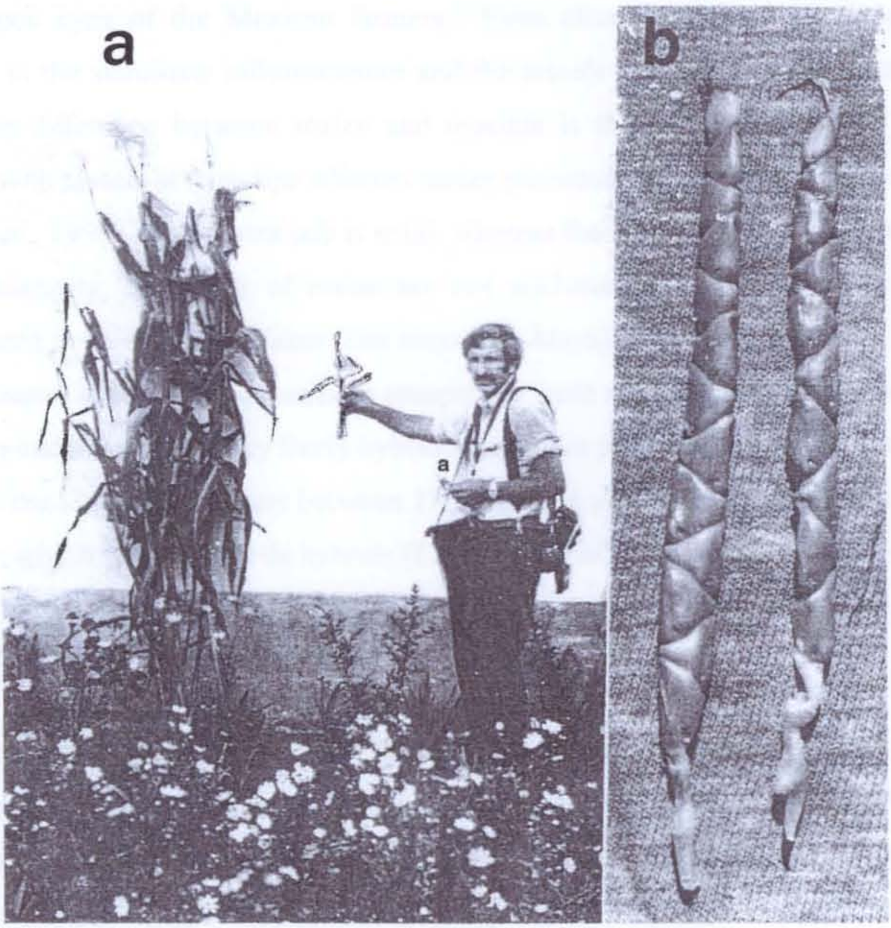

Figure 4.4: The teosinte, *Zea mays* spp. *Mexicana*, in the Valley of Mexico. (a) A robust teosinte plant taken from a maize field and a smaller teosinte plant found growing along the edge of the highway. (b) The ears or female inflorescences of the teosinte *Zea mays* ssp. *Mexicana* which differ remarkably from ears of maize in appearance and structure despite the plants being members of the same biological species (source: Dobley, 1990).

A recent genetic experiment suggests maize (corn) to be more related to annual teosinte than to any of its other relatives (Galiant, 1984). Both have the same chromosome number and they hybridize readily. The fertility of the hybrids is high because their chromosome pairing is regular and virtually complete. Morphologically, teosinte is similar to maize; and indeed when both grow together in the maize fields of the valley of Mexico, distinguishing one from the other before they blossom is not an easy task, even for the keen eyes of the Mexican farmers. Even after flowering there is a marked similarity in the staminate inflorescences and the tassels (Soriano and Klevezas, 2000). The major difference between maize and teosinte is that teosinte typically has long branches with tassels at their tips whereas maize possesses short branches tipped by ears (Wang *et al.*, 1999). The maize cob is solid, whereas the teosinte cob is brittle and come apart at maturity; the seeds of maize are not enclosed, while those of teosinte are encapsulated in fruit cases. Maize (*Zea mays ssp. Mays*) and annual teosinte (*Zea mays* ssp. *Mexicana*) is, however, genetically compatible, both are wind pollinated, and in areas of Mexico and Guatemala they freely hybridize when in proximity to each other (Agbios, 2001). In the US a cross occurs between *Tripsacum* (a genus closely related to Zea) and *Zea mays*, which produce sterile hybrids (Carpenter *et al.*, 2002).

4.1.2.3.2.2 *Cotton (Gossypium Hirsutum L.)*

The cotton genus (*Gossypium L Malvaceae*) consists of approximately 50 species of shrubs and small trees found worldwide in both tropical and subtropical areas (Wendel *et al.*, 1992). It is subdivided into four sub-genera and these further subdivided into sections and subsections. *Gossypium L.* includes four species of cultivated cotton (*G. arboreum* L., *G. barbadense* L., *G. herbaceum* L., *G. hirsutum* L.) (Biotech basics, 2001). *Gossypium hirsutum* in its wild and commercial form grows in the drier areas of Middle America, northern South America, the West Indies, the southern tip of Florida and through introduction in Northern Africa and Southern Asia. The wild population is rare and widely dispersed (Lee, 1984).

At least seven genomes (chromosome sets with distinctive gene groupings) designated A, B, C, D, E, F and G are found in the genus. Diploid species (2n=26) are found on all continents. The A genome is restricted to diploids of two species (*G. arboreum* and *G. herbaceum*). The D genome is restricted in diploids of some species, such as *G. thuberi*. *G. hirsutum* and *G. barbadense* are both allo-tetraploids (plants with four sets of chromosomes derived by doubling of chromosomes from a hybrid plant) (Biotech basics, 2001). *G. tomentosum*, *G. hirsutum*, and *G. barbadense* have compatible genotypes and can be crossed to produce viable offspring, although crosses with *G. tomentosum* are only known with certainty from artificial crosses in breeding programs (Biotech basics, 2001). The bulk of the world's cotton is supplied by modern cultivars of *G. hirsutum* (Fryxell *et al.*, 1991). These four species of Gossypium are/have been widely cultivated in Africa (Vollesen, 1986). Because *G. hirsutum* and G. *barbadense* are sympatric in the Caribbean and co-occur to a limited extent in Central America, it was expected that inter-specific introgression would be most frequently detected (Wendel *et al.*, 1992). *G. barbadense* is also an alien plant in South Africa coming from North Africa. Scientific study proves that cross between *G. hirsutum* and *G. barbadense* can take place successfully (Schendiman *et al.*, 1974). There is hybrid vigour present in F1 hybrid progeny with good productivity, length and strength of the fiber (Ano *et al.*, 1983). This cross occurs mainly because cotton is partially an insect pollinator. Secondly both are tetraploid species and they can cross successfully and give a vigorous F1 hybrid (Hutchinson, 1940). The evidence shows that a cross between *G. barbadense* and Bt cotton can take place in South Africa as they are both tetraploid species and can be pollinated by insects.

The inter-specific crossing between the same genome groups of cotton can produce fertile F1 progeny, but it will segregate and lose its vigor (Harland, 1936; Hutchinson, 1940). Backcrossing with cultivated species can bring back the desirable characteristics (Munro, 1987). In inter-specific crossing the cross may be between a diploid and a diploid plant, or between a diploid and tetraploid plant, giving respectively a diploid or a triploid hybrid. Doubling the chromosome number, giving a tetraploid or a hexaploid plant, can restore fertility. It is usual to try to synthesize such a tetraploid from diploids with the A and D genome, so that it will produce fertile seed when crossed with one of the cultivated tetraploid species (AD) (Munro, 1987). The fertility may not be complete, and may require several backcrosses to the natural tetraploid before full fertility is restored (Munro, 1987).

Under controlled conditions hybridization between plants of *G. hirsutum* and wild *G.*

thuberi would likely result in a triploid (3X=39) sterile plant, because *G. hirsutum* is an allo-tetraploid (4X = 52) and *G. thuberi* is a diploid (2X = 26) plant (LaSota, 1996). *G. herbaceum africanum*, which is a wild form of cotton indigenous to South Africa, is a bushy perennial shrub and is considered to be the most primitive cotton (Munro, 1987). LaSota (1996) showed that a cross between the diploid (2X=26) *G. herbaceum* and allo-tetraploid cultivated cotton could occur, because they are insect pollinated plant, but the cross produces sterile plants.

Chapter 5

Genetically Engineered Plants Grown in South Africa

5.1 Provision of information

5.1.1 Literature search and personal interviews

Beside a literature survey carried out with the use of published material at the university library or on the Internet the following persons contributed with general information about genetically engineered plants to the survey.

J. Webster, Director of Africa Bio, provided references and website addresses that helped to identify the possible risks that are caused by the transgenic maize and cotton in South Africa and the potential benefits that are associated with transgenic crops. The website www.africabio.com provided information about the problems and opportunities of biotechnology for developing countries like South Africa.

S. Moephuli, Director of Genetic Resources at the Department of Agriculture, contributed information concerning the Genetically Modified Organisms Act (GMO Act 15) of 1997 in South Africa.

The website www.biotechnology.gov.au provided by M. Koch helped to find out what the possibilities are for gene flow between weedy relatives and transgenic plants in South Africa. By finding these various possibilities, it allowed to formulate reasonable solutions to reduce the risk of gene flow.

The cotton articles, provided by Dr C.L Bredenkamp, gave information on different types of cotton. More importantly, it gave information on the weedy relatives of cotton in

South Africa, which was used to find out what chance there was of gene flow between transgenic cotton and the weedy relatives. By doing so, a safety precaution was recommended.

The website address www.agbiotechnet.com from Prof. D. Berger contained information related to the risks of transgenic crops, which helped to identify the risk that may be associated with the transgenic maize and cotton of South Africa.

5.1.2 Personal telephone interviews with companies

Personal interviews were also conducted with two seed companies involved in genetically engineered plants to obtain information about the current use and growth areas of these plants. Information obtained from Monsanto/Johannesburg (MC) and Pioneer Hybrid/Centurion (PH) was as follows:

What types of crops are already released commercially?
Maize and Cotton (MC); Maize and Sunflower (PH)

Who are the main customers?
Farmers in Bethel (MC); no answer (PH)

In which Province are the sales of hybrid seeds biggest?
Mpumalanga (MC); Natal (PH)

Do you only sell hybrid maize seeds or do you sell ordinary seeds as well?
Company sells both (MC and PH).

Of these two, which one is of higher demand?
Ordinary seed is of higher demand. Hybrid seeds are of low demand, about 3-5% (MC and PH)

Which is more profitable?

Hybrid seeds (MC and PH)

What are the different traits of hybrid seeds you are selling?

Bt maize, Bt cotton. Round up Ready (RR) soya is already registered, but the seeds are not available. Monsanto also registers RR maize but the seeds are not available (MC).
Bt maize (PH)

Is anyone doing research on the environmental impact on flora?

No idea (MC); Department of Agriculture (PH)

5.2 Result of survey

5.2.1 Current growth areas

The study was concentrated on maize and cotton, as they are the main genetically engineered crops currently grown in South Africa. The overall survey showed that the current use of transgenic hybrid seed by farmers in South Africa is 3-5%. According to the survey, Monsanto's and Pioneer Hybrid's transgenic maize and cotton are mainly planted in the Mpumalanga and Natal region. South Africa is further the only African country in which genetically engineered crops are commercially grown, and it has adopted the technology more quickly than any other country in the world. Already 28% of cotton and 6% of the maize planted in South Africa is genetically engineered (Wynberg, 2002).

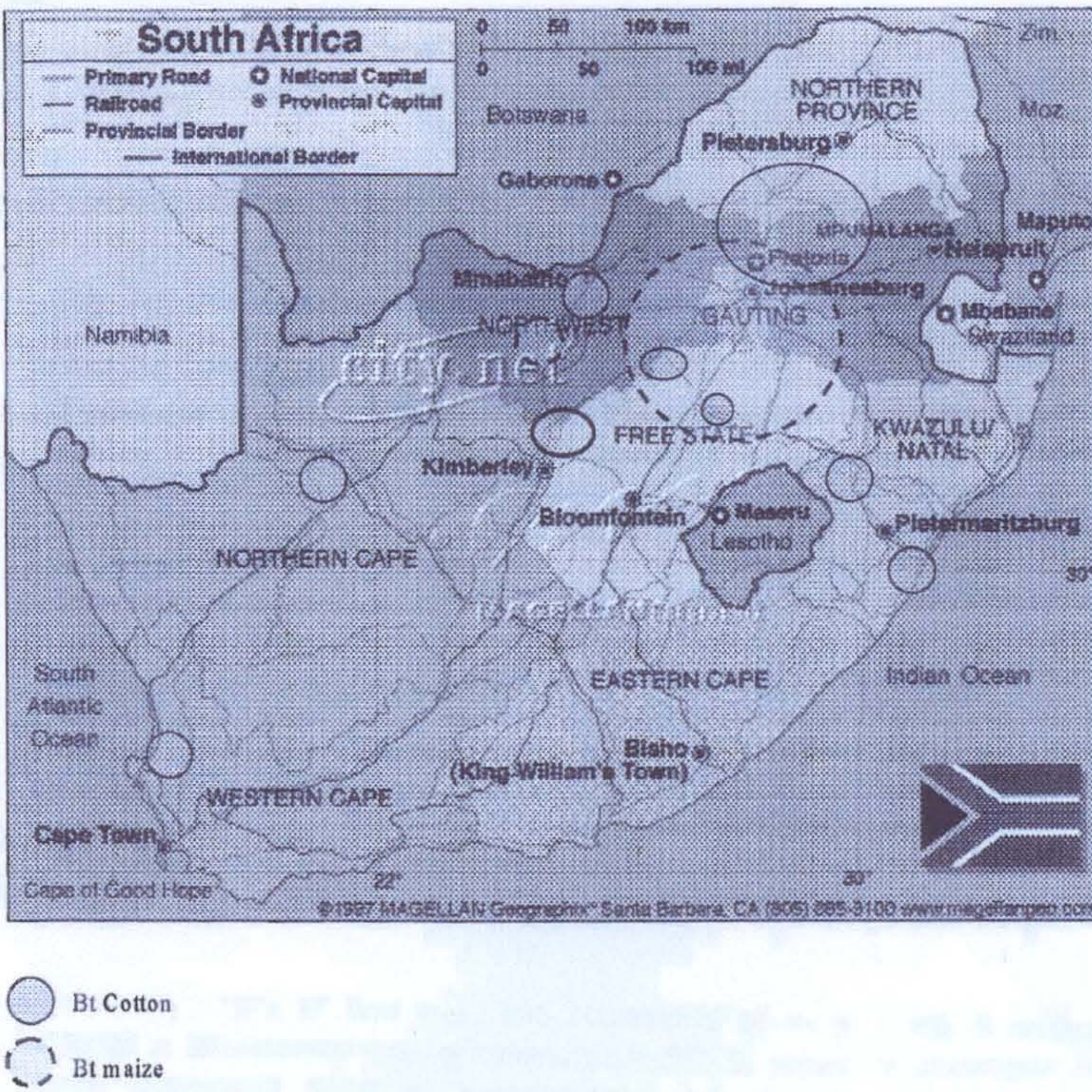

Figure 5.1: Commercial plantings of genetically modified maize and cotton in South Africa (Viall, 2001).

From Figure 5.1 it is evident that Bt cotton is planted in many parts of South Africa. It is also evident that Bt maize is planted within one area covering all of Gauteng and stretching to the North West Province, Free State and Mpumalanga. Around 200 field trials are currently taking place in South Africa and five commercial releases have been approved (Wynberg, 2002). In 2002, Bt cotton has been commercially planted in eight provinces. These are Eastern Cape, Kwazulu Natal, North West Province, Free State, Northern Province, Gauteng, Northern Cape and Mpumalanga. Bt maize is planted in Kwazulu Natal, Eastern Cape, North West Province, Free State, Gauteng and Mpumalanga (Figure 5.2). Field trials of Bt maize are currently conducted in Northern Province and Northern Cape (Figure 5.2). Since 2000, Bt maize was planted in two additional provinces, Kwazulu Natal and Eastern Cape, while Bt cotton was planted additionally in Eastern Cape Province by the year 2002 (Figure 5.1 and 5.2).

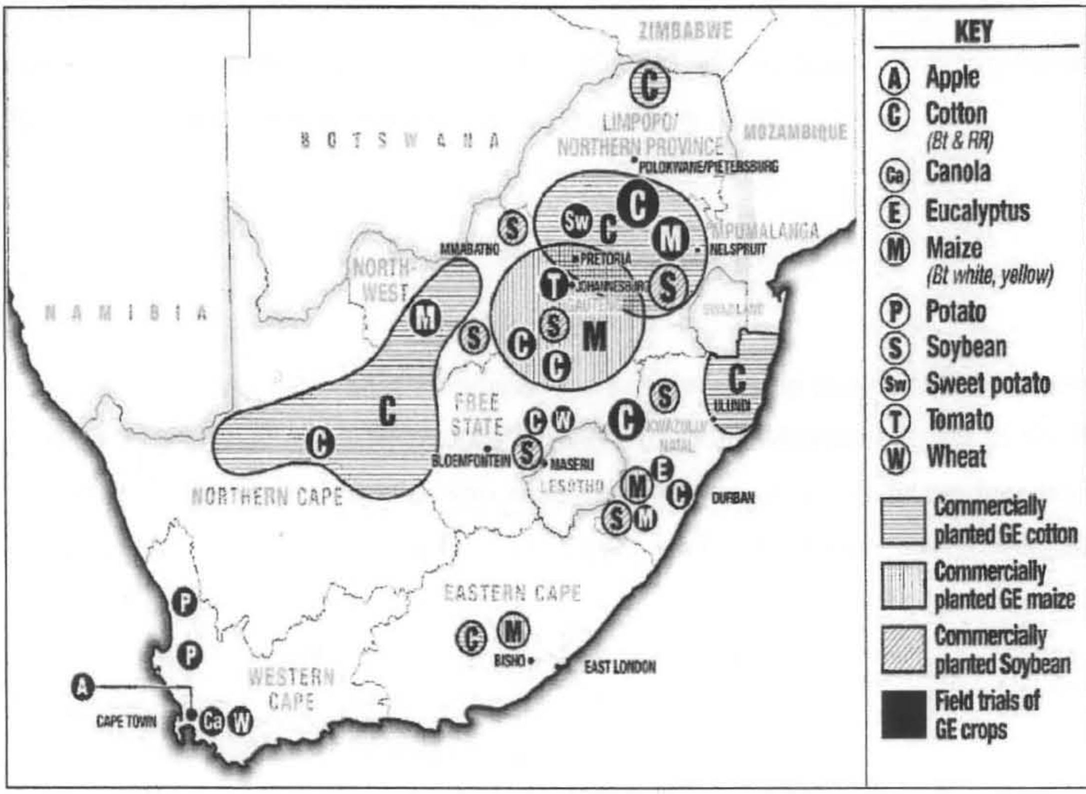

Figure 5.2 Planting of genetically engineered crops in South Africa, 2002 (courtesy of Bio watch South Africa).

5.2.2 Potential benefits

From the interviews carried out, the proponents of genetically modified plants mentioned the following benefits for a developing country like South Africa.

5.2.2.1 *Economic benefits*

The survey showed that those who introduced the transgenic crop benefited from a yield increase of 25% (CropBiotechNet, 2002). Results from the Makhathini flats in South Africa further indicates that farmers who adopted Bt cotton were able to produce high level of output with lesser amount of input, such as labour and chemicals (CropBiotechNet, 2002).

5.2.2.2 *Socio-economic and health benefits*

Even though very poor farmers may not be able to afford GM seeds, the survey on small scale farmers at Makhathini farmers showed that genetically engineered plants might provide the following socio-economic and health benefits:

- Alleviation of hunger and malnutrition in Southern Africa due to increased production of food crops.
- Improvement of the standard of living of the farmers due to drought and insect resistant crops.
- Decrease of farm worker exposure to insecticides and pesticides improving the quality of the environment.
- Bt cotton might give small-scale farmers, mainly women, more time to care for their children and the sick.

5.2.2.3 *Environmental benefits*

The survey further showed that genetically engineered plants might provide the following environmental benefits:

- More efficient land utilization through improved yields, as land is fast becoming a limited resource.
- Promotion of less use of pesticides and herbicides.
- Less need for weed control, fewer passes of machines through the field are needed.
- Reduction of work in the field resulting in less soil compaction with higher oxygen content in the topsoil.
- Reduction in herbicide and insecticide usage reduces the risk of contamination of domestic water sources in rural areas.

5.2.3 Potential risks

From the survey the following two major risks specifically for the flora of a developing country like South Africa were outlined:

- Pollen viability of transgenic crops will determine gene flow between genetically engineered plants and possible wild relatives.
- Gene exchange between varieties of cultivated and genetically modified crops.

5.2.3.1 *Out-crossing with cultivated crops*

The literature survey showed that gene exchange between Bt cotton or maize and cultivated cotton and maize is a risk in South Africa. This is due to the identical ploidy level of species and the planting in close proximity to each other (Figures 5.1 and 5.3). Gene exchange can occur due to wind pollination in cultivated maize and via insect pollination in cultivated cotton. Hybrid production would also pose a serious threat to the environment over time, because any hybrids might be toxic to beneficial insects in addition to bollworm as the target insect. A study conducted at Cornell University in New York conducted by Losey *et al.* (1999) showed that Bt maize pollen may drift onto milkweed around maize fields and can affect the survival of the monarch larvae. According to the laboratory tests, it was found that monarch butterflies feeding on milkweed leaves, which had been dusted with pollen from Bt maize, ate less, grew more slowly and suffered higher mortality than larvae that ate milkweed leaves without any Bt maize pollen. Similar interactions between a cultivated genetically engineered plant and a weed can also not be excluded in South Africa.

Continuous planting of genetically engineered plants might also increase herbicide resistance not only in cultivated plants but also in closely grown weeds. For example, a continuous usage of glyphosate in genetically engineered maize and cotton could increase the resistance for this herbicide in non-crop plants associated with these two crops and promote the development of 'super weeds'. This would require the use of a higher herbicide dosage for weed control and might reduce yield because of competition with the new 'super weed'. An increase in herbicide resistance due to the use of genetically engineered plants has been already found in the U.S. (S. Duke, personal communication). Such a possibility should also be considered in South Africa as a longer-term effect on the environment.

5.2.3.2 *Out-crossing with wild relatives*

5.2.3.2.1 *Maize*

The survey showed that there is no sexually compatible wild or weedy relative of maize that is currently known in South Africa. Maize has only one related wild species, teosinte, which grows only in Mexico and Guatemala but not in South Africa. Thus, concerning gene flow to wild relatives by out-crossing, genetically engineered maize can be considered as relatively safe for South Africa.

The literature survey also showed that some relatives of corn are wild plants but with no pronounced tendency for weediness (Galiant, 1984). In the U.S., but not in South Africa, a cross occurs between *Tripsacum*, (a genus closely related to Zea) and *Zea mays*. But resulting hybrids are often sterile, because of difference in chromosome number and lack of pairing between chromosomes (Eubanks, 1997; Carpenter *et al.*, 2002). A herbicide-resistant character, however, might create volunteer transgenic plants in crop rotation fields, which might also be relevant to South Africa. If viable genetically engineered maize seeds are lost in the harvesting process, weediness will become a problem as volunteer weeds reduce yield. Maize also appears as a volunteer at roadsides, but it has never been able to establish itself outside of cultivation.

From the literature survey it is also evident that the hypothesis that escape of volunteer corn plants will have a selective advantage and become weeds is empirically unfounded. This has been clearly dismissed by the results of an intermediate term experiment conducted in the UK. In the experiment, oilseed rape, potato, maize and sugar beet were grown in 12 different habitats and monitored over a decade to find out whether genetically engineered plants were likely to persist in the wild in the event of dispersal from their cultivated habitat. The results showed that plants were no more invasive or persistent than their conventional counterparts (Crawly *et al.*, 2001).

Due to the difference in climatic conditions between South Africa and the UK, a study should be conducted in South Africa to find out whether volunteer weeds become persistent or invasive in the natural environment.

5.2.3.2.2 *Cotton*

The literature survey showed that among the African species (Figure 5.3), *Gossypium triphylluim* is closely related to *G. anomalum* and artificial hybrids show a certain degree of fertility (Saunders, 1961). But where the two co-exist (the border area between Angola and Namibia), natural hybrids have never been recorded (Fryxell, 1980). *Gossypium arboreum* (diploid; 2n) is one of the four domesticated *Gossypium* species and is the only one for which a wild progenetor has never been identified (Liu *et al.*, 2001). *G. annomalum* also called WaWra and Peyr is a wild diploid cotton indigenous to the African continent. This species and G. *somalense* have been collected in natural vegetation in the Sudan (Saunders, 1958).

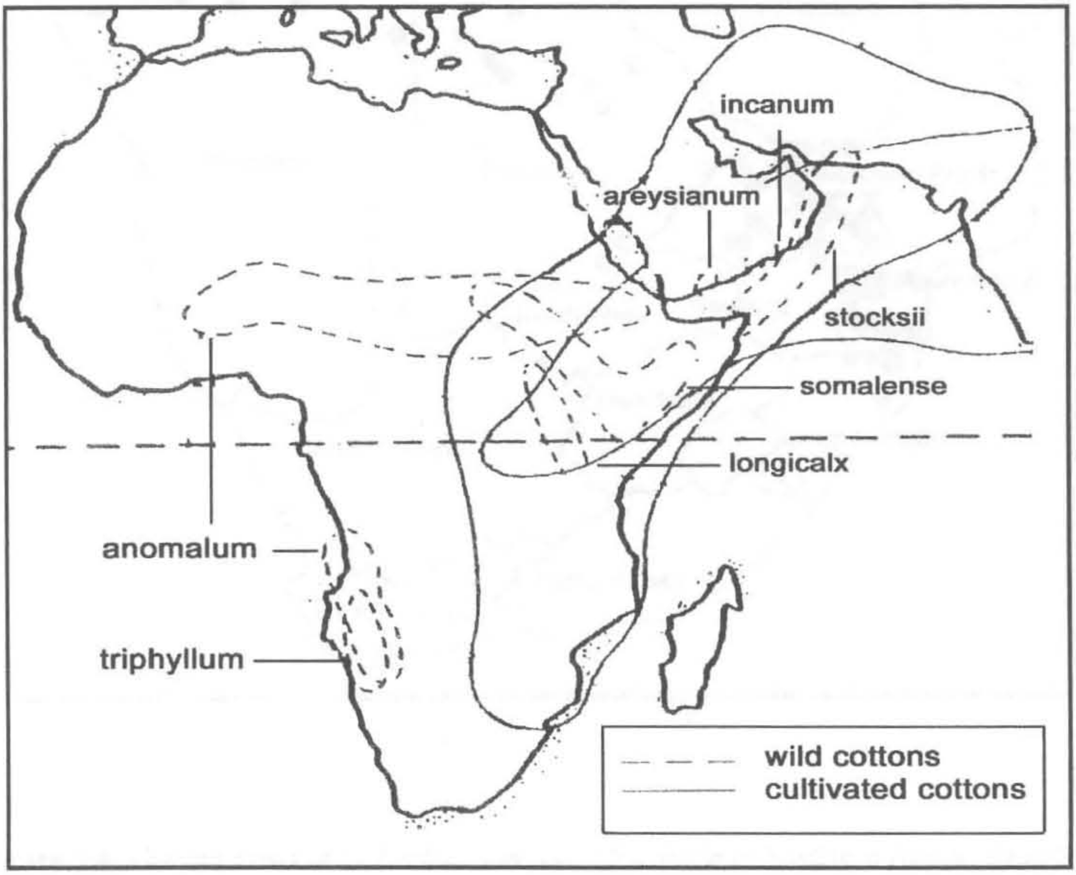

Figure 5.3: The natural geographic distribution of *Gossypium* species in Africa (Valicek, 1978).

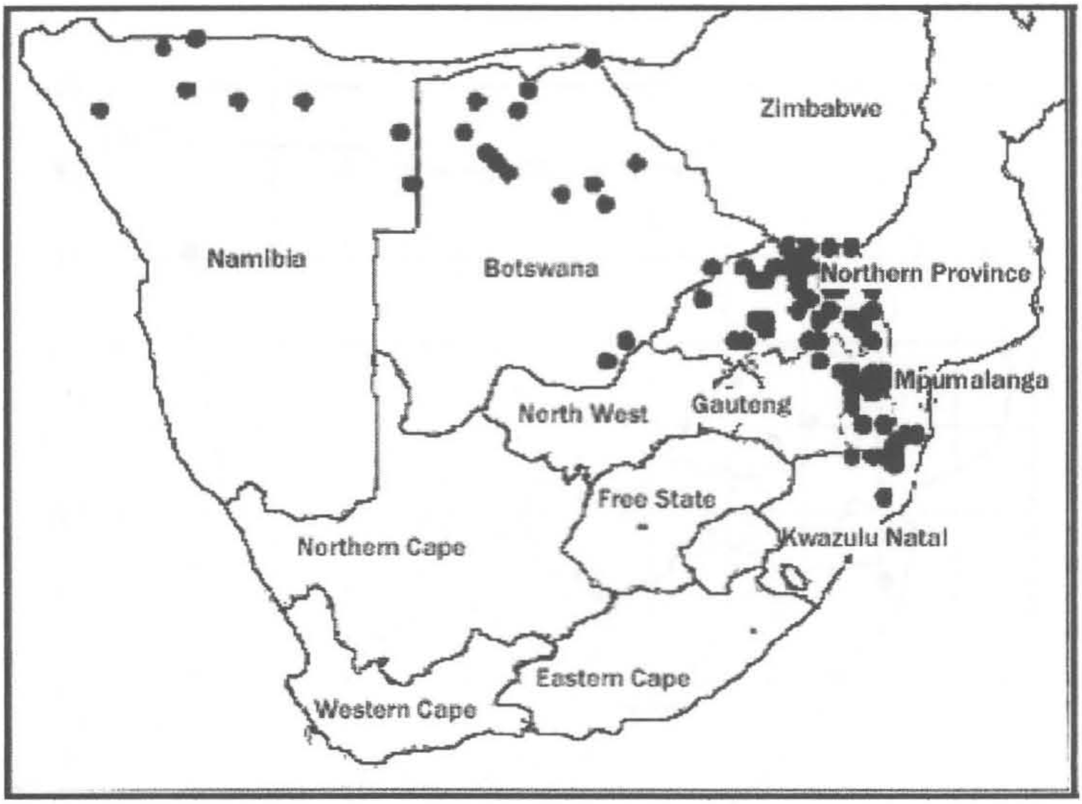

Figure 5.4: Growth areas of *G. herbaceum* race *africanum* in Southern Africa (Saunders, 1961).

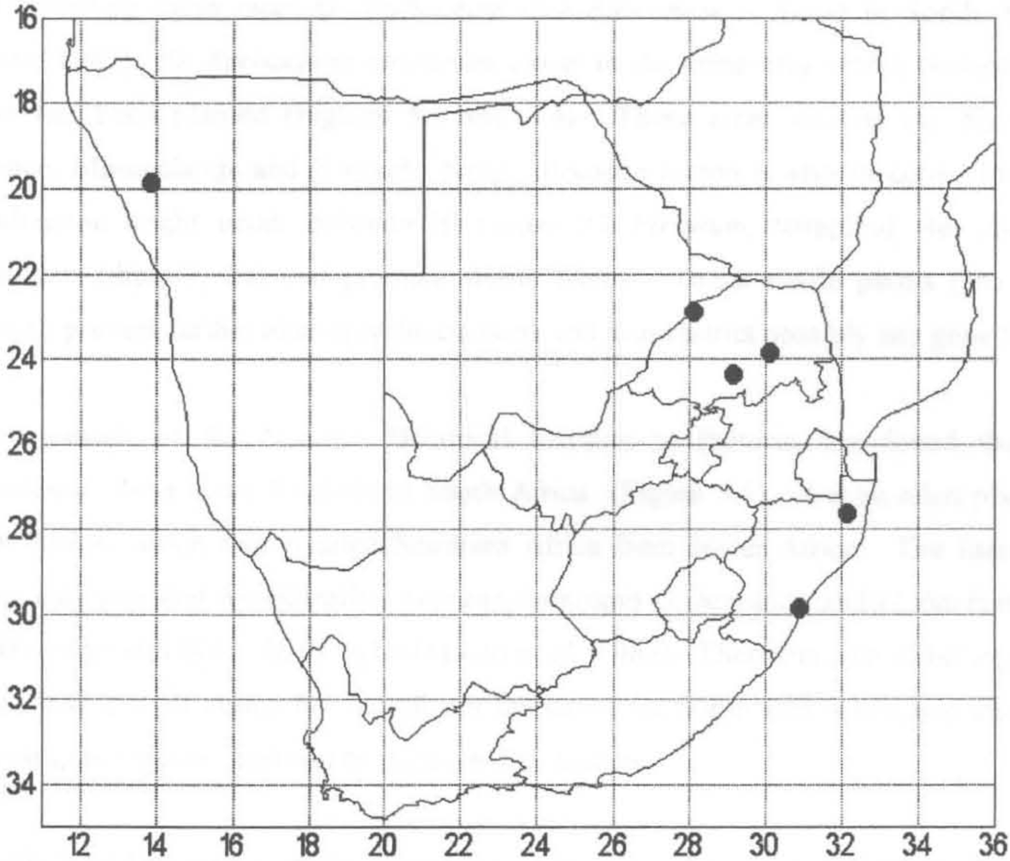

Figure 5.5: Growth areas of *G. barbadense* in Southern Africa, 1999 (map obtained from National Botanical Institute, Pretoria, South Africa).

G. herbaceum is divided into five geographical races, which are now isolated from each other. Among these races *G. herbaceum* race *africanum* is found in South Africa (Munro, 1987). *G. herbaceum africanum* exists in the same area where currently Bt cotton has been planted (Figures 5.1 and 5.4). These areas include the Northern Province, Mpumalanga and Kwazulu Natal. Because cotton is also insect-pollinated, hybridization might occur between Bt cotton (*G. hirsutum*, tetraploid; 4n) and *G. herbaceum* (diploid; 2n) and produce sterile plants. These sterile plants (3n) will, however, prevent further inter-specific crossing and thus restrict possibly any gene flow.

A data search, at the National Botanical Institute in Pretoria, has found that *G. barbadense* (Pima cotton) occurs in South Africa (Figure 5.5). It is an alien plant in South Africa, which has invaded Southern Africa from North Africa. The literature survey indicates that hybridization between tetraploid *G. hirsutum* and *G. barbadense* occurs and produces fertile F1 hybrids (Ano *et al.*, 1983). Therefore a cross between *G. barbadense* and Bt cotton has significant relevance for South Africa because they are both tetraploid species and can be pollinated by insects.

G. thuberi Todaro occurs in the mountains of Southern Arizona and Northern Mexico (Wozniak, 2002). There is evidence of hybridization between *G. hirsutum* (4X=52) and *G. thurberi* (2X=26) to produce triploid (3X=39) sterile plants (LaSota, 1996). Even though there is no *G. thuberi* in South Africa the above evidence shows that a cross between diploid *G. herbaceum* and Bt cotton can take place to produce sterile plants.

Chapter 6

Discussion and Conclusion

Genetic modification of plants can be exploited to meet the ever-growing need for food and novel plant-based products. But as with any new scientific field, there are many concerns about the possible risk of such genetically modified crop plants. These concerns have to be addressed allowing the production of a safe and environmentally friendly crop. Although such plants have undoubtedly economic benefits there are major concerns about the risks such plants might pose for the environment. This study has therefore tried to identify such possible risks on the South African flora and to highlight areas that need attention.

6.1 Environmental risks

Would a herbicide-resistant genetically modified plant pose a risk in South Africa? Resistance gene flow from a genetically modified plant is regarded as a typical risk. Several research groups have already recognized this undesirable consequence of gene flow (Reiger *et al*., 1999; Dale, 1992; Ellstrand, 1988). In the case of a herbicide-resistant weed, such a weed would be difficult to control. It might develop resistance against most potent herbicides that are currently used for its control (Carpenter *et al*., 2002). However, any transfer of an introduced herbicide resistance gene to a wild plant would only be effective when this plant is treated with the respective herbicide. Without such herbicide treatment, the resistance gene will be integrated into the host genome but without any further obvious consequence for the plant itself or its progeny.

Would herbicide-resistant maize pose a risk in South Africa? Maize does not have any sexually compatible wild or weedy relatives in South Africa with which out-crossing can occur. Teosinte, the closest wild relative of maize, grows only in Mexico and Guatemala but not in South Africa. Any risk of dispersal of any introduced gene into maize to a weedy relative does therefore not exist. Consequently, planting of genetically modified maize will not pose any significant risk to the South African flora regarding gene flow to wild relatives. This would be, however, in contrast to cotton. Cotton has wild or weedy relatives in South Africa and poses a certain degree of risk like transgene flow and harmful effects on non-target organisms.

Is gene flow to a cultivated crop a risk in South Africa? Gene flow from genetically modified maize can occur to non-modified maize under cultivation in South Africa. However, any gene flow between commercial varieties should be regarded more as a legal issue than as an environmental risk and any risk on the flora is negligible. As maize is wind-pollinated any out-crossing results in the production of fertile maize hybrids. Hybridisation significantly affects seed quality and is therefore not desirable (Agbios, 2001). Also, to control undesired hybrids, which might carry a herbicide resistance gene, a greater amount of a herbicide has to be applied for their control with the building up of resistance towards the herbicide and also a higher level of pollution of the environment. Generally, the resistance gene might disappear after five generations in case no herbicide is sprayed for plant control.

Does the creation of super weeds and volunteer plants pose a risk in South Africa? A genetically modified plant might turn into a super-weed and increase the chance of plant invasion. There is, however, evidence that the direct effect of an introduced herbicide resistance gene may be neutral with regard to fitness in natural environments as long as the herbicide is not applied (Raybould and Gray, 1994; Duke *et al.*, 2002). Crawley *et al.* (2001) found no evidence for enhanced weediness in genetically modified *Brassica napus* expressing a herbicide resistance gene. In South Africa, however, the chance of super-weed creation as a risk for the South African flora is extremely limited due to the lack of practise to apply herbicides on natural plant populations. This is also true for volunteer plants carrying as genetically modified plants multiple-herbicide resistance. This is a clear threat to commercial farming, including organic farming, but does not pose a threat to the flora as long as herbicides are applied for selection.

Are insect-resistant plants a risk for the South African flora? While herbicide-resistant plants generally pose a limited risk, the study identified an obvious risk for genetically modified plants carrying a gene for insect resistance, such as the Bt gene. This is specifically true for genetically modified cotton. Any gene flow and expression of the Bt gene in genetically modified plants and their wild relatives would affect both target and non-target insects feeding on both types of plants and would therefore also indirectly affect natural predators and pollen transfer by insects. In South Africa, the majority of Bt cotton is planted in Mpumalanga and North-eastern Kwazulu Natal. Gene flow from genetically modified Bt cotton (*G. hirsutum*; tetraploid) to wild cotton (*G. herbaceum*, diploid) when grown in close proximity is possible. But such a cross would very likely produce triploid sterile plants. However, as the study also showed, *G. barbadense* (tetraploid), which is an alien to South Africa but invaded South Africa from Northern Africa, has been sporadically found in the past in Natal and the Limpopo Province in close proximity to areas where Bt cotton is planted. Hybridization between *G. barbadense* and *G. hursutum*, which are both tetraploid, is possible through insect pollination. It produces a viable, fertile hybrid population with a hybrid vigor (Wendel *et al.*, 1992, Ano *et al.*, 1983; Schwendiman *et al.*, 1974) and back-crossing with one of the parents results in hybrid stabilisation (Munro, 1987). Consequently, there is a risk that in South Africa gene exchange might occur between tetraploid cotton due to insect pollination especially when planted in close proximity. In South Africa, the distance between Bt crop fields and *G. barbadense* is 30 km, which significantly reduces the risk of any gene flow. Pollen remains viable for about 12 hours, after this period the pollen looses its viability, thus the longer it takes for the pollen to reach a relative, the less the chance of out crossing (Govila *et al.*, 1969; Banks, 1998).

In South Africa, a further general risk for plants with a wild relative is that an adaptive resistance gene might enter a related wild population giving the progeny of some individuals a large competitive advantage. The genotype of these could, by "genetic hitch-hiking", sweep the population, eliminate other genotypes, and reduce the amount of genetic variation. This could also have practical implications when genetically modified plants are widely applied in South Africa, because important genes would be eliminated from a natural "germplasm bank" (Regal, 1994). For example, in the US more than 80% of seed varieties sold a century ago are no longer available. Less than 20% of vegetable seed varieties listed in a 1904 US national inventory are still commercially available today. Scientists have also warned that "genetic pollution" of Mexico's many maize varieties could lead to the loss of the world's most important and irreplaceable source of corn germplasm (World watch Institute, 2000; Cummins, 2002).

6.2 Administrative risks

Beside environmental risks, the study also identified as a risk the still existing inadequate expertise in South Africa for assessing risk of genetically modified plants including control and supervision of the GMO Act. The Executive Council set up under the GMO Act is not all-inclusive. It excludes scientists from the research institutions in the actual decision making process. From the literature survey it is evident that no comprehensive risk assessment study has been so far conducted on the eco-system and vulnerable species. As an immediate action, extensive training in bio-safety and risk assessment should be carried out. Further, despite the fact that the Act calls for an assessment to be conducted on both the environmental risk and the risk it may have on human health, the Act does not specify the basic standard as to what is an acceptable risk. There is also no provision for public participation in various structures designed by the GMO Act.

6.3 Actions for risk limitation

What type of actions should be considered to limit the current risks on the South African flora? Through risk limitation strategies gene flow can be reduced considerably. For example, this includes simple cultural practices. Crops might be planted so that they do not flower at the same time as their wild relatives or complete harvesting of an annual crop before flowering will prevent pollen formation and possible pollen transfer to wild relatives (Ellstrand and Hoffman, 1990). One interesting scientific strategy to prevent out-crossing is also to insert new genes into the chloroplast of the cell rather than the nucleus. Since the chloroplast genomes are not passed through the pollen, they will not transfer any introduced gene into the chloroplast (Brookes, 1998).

What further strategies could be followed to limit any gene flow from genetically modified plants to wild relatives in South Africa? From the literature survey 'distance isolation' can be considered as an important aspect to prevent gene flow to wild relatives. Results for the importance of distance isolation have been obtained from experiments investigating the effectiveness of isolation distance (Luna *et al.*, 2001). Most self-fertilizing species including cotton, which is only partially pollinated by insects, require isolation distances of more than 200 m. In contrast, out-crossing species require 1000 m or more for isolation (Ellstrand and Hoffman, 1990). A recent study completed at the University of Maine found that cross-pollination by conventional maize, a wind-pollinated plant, with genetically modified maize in an adjacent plot was 1% at a distance of 30 m and declined to zero at a distance of 300 m. This suggests that it is possible to drastically limit the transfer of a gene from genetically modified to non-modified plants by following the same recommended planting distances currently in place to maintain purity in conventional plant varieties (Colorado State, 1999).

Insect pollinators, primarily bumblebees and honeybees, are agents for pollen dispersal in the cotton growing regions of the USA. An isolation distance of at least 400 m is required from other cotton to avoid insect pollination (Sumida, 1995). Further, buffer zones might prevent cross-pollination. A buffer zone is an area of non-genetically modified crops to prevent pollen drifting into nearby fields and pollinating other crops and weeds. It acts as a trap for pollen carried from the genetically modified plants by insects such as bees. Bees are most likely to visit the flowers on the buffer plants further away (CSIRO Australia, 2000). Estimates of the necessary width of buffer zones vary depending on the genetically modified crop. For example, cotton pollen has a low tendency to drift around, requiring only a small buffer zone. But for crops like maize where wind dispersal of pollen occurs, much larger buffer zones are required (CSIRO Australia, 2000). South African farmers seemingly maintain the recommended safety distance between the transgenic and cultivated crops (C. Laubscher, personal communication) reducing the risk of gene flow.

To limit any risks in South Africa, a tighter safety regime should also be established for genetically modified crops. This should be applied specifically to genetically modified plants with an obvious impact on the environment, such as Bt cotton expressing a toxin. It should also cover tightly controlled field trials in the country carried out by independent scientists to evaluate the short and long-term impact on the ecology under natural conditions. This is important, because the result under natural conditions may be different from the result under laboratory conditions. Estimation of risk should be done on a crop-by-crop basis in the region where the modified crop is to be cultivated and not only be based on experiences in different countries with different environmental conditions and ecology.

6.4 Conclusions

In general, there should be a more cautious approach in regard to the use and release of genetically modified plants in South Africa due to its possible impact on the environment. Although currently known risks in South Africa are rather minimal, several unknown risks might include potential ecological and human health risks that have not been studied in greater detail in South Africa. So far, various genetically modified crops have already/or will be released in South Africa including genetically modified maize and cotton, with soybean to follow and South Africa is among the few countries in the world rapidly introducing engineered crops. There is certainly a risk, which requires urgent attention, when a technology is deployed too fast without sufficient safeguards, regulations or public debate and proper risk assessment studies.

Chapter 7

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Annex

Methods

The following search methods have been applied to obtain primary data for the impact assessment on genetically engineered plants:

Questionnaire

To acquire primary data, a questionnaire was designed, which was applied to obtain specific information about transgenic crops used in South Africa. Answers to the questionnaire were obtained by personal interviews or by telephonic conversation. The questionnaire consisted of the following questions:

- What types of crops are already released commercially in South Africa?
- Who are the main customers?
- In which Province are the sales of hybrid seeds biggest?
- Do you only sell hybrid maize seeds or do you sell ordinary seeds as well?
- Of these two, which one is of higher demand and which is more profitable?
- What are the different traits of hybrid seeds you are selling?
- Is anyone doing research on the environmental impact of genetically engineered crops on the South African flora?

Personal Interviews

To obtain information about genetically engineered plants and possible environmental risks involved of growing genetically engineered plants the following persons were contacted in South Africa:

J. R. Webster: Executive Director of AfricaBio. Promotes research, development and application of biotechnology. Provided useful references and contact addresses. Helped to find out the companies in SA selling transgenic seeds

S. Moephuli: Director, Genetic resources, Department of Agriculture. Expertise in Biochemistry. Provided information about the Genetically Modified Organisms Act. The objective of the act is to promote the responsible development, production, use and application of GMOs. Helped assessing the existing bio safety regulations.

M. Koch: Director of innovation technology, Secretariat to the South African committee for genetic experimentation (SAGENE). Provided useful information and guidelines for the preparation of the questionnaire for the general release of genetically modified plants. Gave some website addresses that were very useful to find out the relevant information.

C.L. Bredenkamp: Assistant curator, National Botanical Institute, National herbarium Pretoria. Provided various literatures about cotton to find different species of cotton in South Africa.

C. Van Gneewe: Monsanto, conducting research, development and marketing of GM seeds. Responded to the questionnaire. Through his response locations in South Africa were identified where transgenic crops are currently planted.

J. Prinsloo: Agronomist, Pioneer Hybrid; Conducting research, development and marketing of seeds. Responded to the questionnaire and provided information on different traits of GMOs the company is selling.

Dr. Dave Berger: Lecturer, Department of Botany, University of Pretoria. Provided useful information about transgenic plants and website addresses.

Internet information

Database search

Most of the information obtained for methods of risk assessment was done by collecting secondary data visiting on the Internet the following databases:

- Ovid (http://gateway.ovid.com)
- Cambridge Scientific Abstracts (CSA)(www.csa2.com)
- AgbiotechNet (http://www.agbiotechnet.com)
- AgBioView (http://www.agbioworld.org)
- AfricaBio (www.Africabio.com)
- Agbiosafety (http://www.agbiosafety.unl.edu)
- CropBiotechnet (http://www.isaaa.org/kc/)

The following key words were used for the searches:

- Environmental impact
- Transgenic plants
- Risk and transgenic plant
- Herbicide resistance
- Gene Transfer

Website search

The following websites gave valuable information for carrying out a risk assessment for the impact of genetically engineered plants on the environment.

- http://www.agbios.com: AGBIOS is a Canadian company dedicated to providing public policy, regulatory, and risk assessment expertise for products of biotechnology.

Search Words: Gene transfer, weediness, herbicide tolerance

- http://www.biotech-info.net: This site covers all aspects of the application of biotechnology and genetic engineering in agricultural production and food processing and marketing.

Search Words: Environmental impact, Herbicide tolerance and insect resistance

- http://www.isaaa.org/: ISAAA is a small, responsive, non-bureaucratic international network with centers in developing and industrialized countries.

Search Words: Biotechnology problems and opportunities, Global status transgenic crop crops.

- http://www.colostate.edu: This is the homepage of Colorado State University; this site contains information conducted by their students on biotechnology. Discusses risks and concerns of transgenic products.

Search Words: Transgenic crops, current transgenic crops.

- http://www.cgiar.org: The CGIAR web site is a collaborative effort of many individuals. This site contains articles agricultural biotechnology.

Search Words: Biotechnology Developing countries.

- http://genetech.csiro.au/: The CSIRO site aims to provide scientific information about gene technology to the public.

Search Words: biotechnology benefits & risks

- http://www.beyonddiscovery.org/content/view.article.asp?a=167: This site contains detailed information on the genetic engineering of plants.

Search Words: transgenic plant, transformation techniques

- http://www.agbiosafety.unl.edu/: This site is maintained at the University of Nebraska-Lincoln with funding from the Council for Biotechnology Information, an industry group. Of special note is the database of released transgenic crop varieties, with details on the genetic modifications and extensive information on food and environmental safety evaluations of each variety.

Search Words: gene transfer and weediness.

- www.agbioworld.org/: Brings the information about technological advances in agricultural in the developing world.

Search Words: 'biotechnology' and 'developing countries'. 'Biotech and Africa'

- http://www.agbioforum.org/: Publishes articles on scientific, economic, and public policy aspects of agricultural Biotech.

Search Words: biotechnology future benefits

- www.monsanto.com: Monsanto Company is a leading global provider of agricultural products and integrated solutions that bring together chemicals, seeds, and biotechnology traits to improve farm productivity and food quality. This site contains information about transgenic plants in various parts of the world and it has a good discussion group.

Search Words: Transgenic plant, Environmental safety

- www.syngenta.com: Syngenta is a world-leading agribusiness. The company ranks first in crop protection, and third in the high-value commercial seeds market. This site contains information on the protection of a variety of crops

Search Words: Herbicide benefits

Internet discussion groups

To obtain personal views on risks caused by genetically engineered plants on the flora the following discussion groups were joined:

- Monsanto Online Discussion Group (UK & Africa)
  www.monsanto.co.uk & www.monsantoafrica.com

- Your Child Nutrition Source: Biotechnology Online Chat
  http://www.asfsa.org/continuinged/onlineed/chats/chat102501.asp

Library information

A further source for secondary data was a thorough search of available books in the library of the University of Pretoria for the following information:

- Methods for risk assessment
- Herbicide resistant crops
- Plant genetic engineering
- Environmental Impact of transgenic crops.
- Transfer of engineered genes to wild relatives.

Maps were obtained from the National Botanical Institute in Pretoria.

Data storage

All Internet and library information on genetically engineered plants were stored in a ProCite database (Windows version 4.01 from Research Information Systems Carlsbad, CA 92009-1572 USA; http://www.risinc.com) allowing a query search. The personal database was screened for the relevant information and extracted information was used for preparing the thesis.