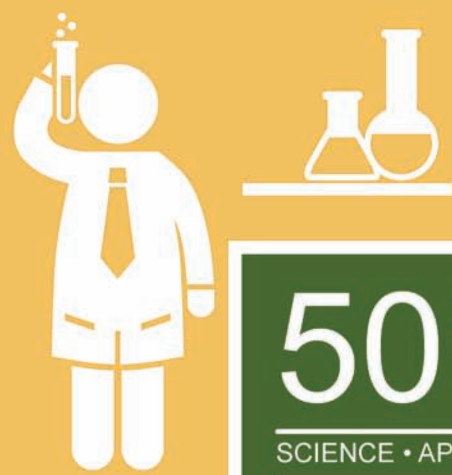

50 BIOTECH BITES

SCIENCE • APPLICATIONS • ISSUES

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Preface

Biotechnology is a set of tools that uses living organisms (or parts of organisms) to make or modify a product, improve plants, trees or animals, or develop microorganisms for specific uses. A variety of biotechnology tools is available that includes conventional plant breeding, tissue culture technology, plant disease diagnostics to more modern techniques such as genetic engineering, molecular breeding and marker-assisted selection, and nanotechnology. Scientists continue to develop several applications and products that are contributing to alleviation of poverty and hunger.

For the last 15 years, ISAAA has been releasing a series of short publications on biotechnology packaged in a form called *Pocket Ks* (Pocket of Knowledge). Its format initially conceptualized as short, concise information fit to put into a pocket has since been reformatted to optimize reading on PC or mobile devices.

These PKs, as they are also referred to, have been translated into other languages such as Bahasa Indonesia, Bengali, Thai, Urdu, and Vietnamese. Their translations have enabled a wider reach for these materials targeting students, teachers, policy makers, media practitioners, and practically anyone who wants more details and reference-based information. Organizations reprint selected issues and disseminate these during biotech events, seminars, and workshops. They have also been used as references for articles and media-based projects such as essay writing, video and cartoon projects on biotech. These PKs are also among the most downloaded materials on www.isaaa.org attesting to their usability and relevance at a time that readers are looking for authoritative, easy to ‘bite’ chunks of information.

50 Biotech Bites is a consolidation of 50 topics, now made available in a single, ready to use publication and categorized into themes. Majority of the series have been peer-reviewed, provided references for further reading, and updated when possible. Former and present staff of ISAAA, particularly the Global Knowledge Center on Crop Biotechnology (KC), did the literature review, wrote the articles, and coordinated with experts. May this publication 'feed' those eager to know more about biotech so that they can engage with others on the technology and make informed decisions about its adoption and use.

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Biotechnology Tools

Conventional Plant Breeding

Conventional plant breeding has been around for hundreds of years and is still used today. Since agriculture began, farmers have been altering the genetic makeup of the crops they grow through selection of the best plants and seeds and saving them for the next season. Early farmers also discovered how some plants can be cross-pollinated to combine the desirable characteristics of the parent plants in their offspring.

Recognizing desirable traits is vital in plant breeding. Breeders scrutinize their fields in search of individual plants with desirable traits. As plant breeding is further developed, breeders understand better the selection of superior plants and use them to create new and improved varieties. These activities have dramatically changed the productivity and quality of domesticated plants.

Hybrid seed technology

Plant breeding can result in either an open-pollinated (OP) variety or an F1 (first filial generation) hybrid variety. OP varieties, when maintained and produced properly, retain the same characteristics when multiplied. The only technique used with OP varieties is the selection of the seed-bearing plants.

Hybrid seeds have improved qualities, such as good vigor, trueness to type, heavy yields and high uniformity, compared to open pollinated varieties. Other characteristics such as earliness, disease and insect resistance and good water holding ability have also been incorporated into most F1 hybrids.

Hybrids are developed through hybridization or crossing of 'pure lines' — plants which, when self-pollinated, produce offspring that closely resemble the parents. By crossing pure lines, a uniform population of F1 hybrid seeds can be produced with predictable characteristics.

For example, a plant breeder picks a plant with good traits but needs improvement on a certain trait. The breeder then picks another plant with not necessarily good overall traits but has the desirable trait to be improved. Each plant will be self-pollinated separately. Their offspring seeds will be harvested and then re-sown. The procedure is repeated for several seasons until identical plants appear each time the seeds are sown. These are now "pure lines".

The breeder now takes each pure line and cross pollinates them. The result is known as an "F1 hybrid." Plants are grown from the seed produced, and the result of this cross pollination should have the combined traits of the two parents.

However, creating F1 hybrids involves years of preparation. A pure line takes seven to eight years to develop before being used in hybridization. Furthermore, to ensure that no self-pollination takes place, all hybridizations of the pure lines are often done by hand.

Another disadvantage is that the plants grown from seeds of F1 hybrids do not perform as well as the F1 material. This requires the farmer to purchase new F1 seeds from plant breeders each season. The farmer is, however, compensated by higher yields and better crop quality.

Though more expensive, hybrids have had a huge impact on agricultural productivity. Today, majority of all corn and rice are hybrids. In the U.S., the use of corn hybrids has more than tripled corn grain yields. Hybrid technology also helped China increase its rice production. Research at the International Rice Research Institute (IRRI) in the Philippines and other countries also prove the potential of hybrid technology in increasing rice yield.

Many cultivars of popular vegetables or ornamental plants are also F1 hybrids. Tropical vegetable breeders have improved plant characteristics over the last two decades including:

- **Yield improvement.** Hybrids often outyield traditional OP varieties due to its improved vigor, improved genetic disease resistance, improved fruit setting under stress, and higher female/male flower ratios.

- **Extended growing season.** Hybrids often mature earlier than local OP varieties. For many crops, hybrid's advantage over OP is most pronounced under stress conditions.

- **Quality improvement.** Hybrids have helped stabilize product quality at a higher, and more uniform level.

Mutation breeding

Occasionally, good traits also arise spontaneously through a process called mutation. However, it is unreliable in terms of producing plant traits that breeders want. Moreover, the natural rate of mutation is very slow.

However, in the late 1920s, researchers discovered that they could increase the number of mutations by exposing plants to X-rays and chemicals. Mutation breeding was further developed after World War II, when the techniques of the nuclear age became widely available. Plants were exposed to gamma rays, protons, neutrons, alpha particles, and beta particles to see if these would induce useful mutations.

Mutation breeding efforts continue around the world today. The International Atomic Energy Agency (IAEA) holds a record of 3,222 varieties approved through mutation breeding. Plants that have been produced via mutation breeding include wheat, barley, rice, potatoes, soybeans, and onions.

Conclusion

Conventional plant breeding has had a huge impact on agricultural productivity over the last decades. However, conventional plant breeding also has limitations. First, breeding is only possible between plants that can sexually mate with each other. This limits the traits that can be added to a particular species. Another limitation is that other traits, including undesirable ones, are also transferred along with the trait/s of interest, which may affect yield potential.

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Tissue Culture Technology

Scientists have searched for methods to make exact copies of superior plants that possess good traits. However, plants usually reproduce through sexual reproduction where sex cells, containing DNA, combine unpredictably, creating unique plants.

This unpredictability presents problems for plant breeders as it lengthens the time required for breeding plants. However, researchers have developed methods of making exact copies of plants through “tissue culture”.

What is tissue culture?

Tissue culture (TC) is the cultivation of plant cells, tissues, or organs on specially-formulated nutrient media. Under the right conditions, whole plants can be generated from a single cell using this technology.

Tissue culture has been vital in the production of uniform, disease-free, quality plants and planting materials. Micropropagation, a tissue culture technique enabling the production of multiple copies of plants relatively quickly, is used to multiply planting materials for large scale planting. Micropropagated plants establish quickly, grow more vigorously, have shorter and more uniform production cycle, and yield higher than conventional propagules.

Tissue culture only requires a sterile workplace, nursery, and greenhouse, and trained manpower. However, it can be labor intensive, time consuming, and costly. Important crops have been grown using tissue culture such as banana, rubber, sweet potato, and tomato.

Uses of TC in Asia

- Tissue culture is used for orchids and hybrids in Southeast Asia where ornamental and cut flower trade is a significant enterprise.
- Thailand, the world's leading exporter of orchids, benefits from tissue culture to reproduce slow-growing and environment-sensitive orchids.
- To control viral diseases, the Philippines uses micropagation for mass propagation of banana.

Benefits of TC for small-scale banana producers in Kenya

Kenya experienced a decline in banana production in the last two decades, mainly due to soil degradation as well as pest and disease infestation and made worse by propagation using infected suckers. With the situation threatening food security and the economy of banana-producing areas, tissue culture was considered as an option to provide enough quality planting materials. With proper management and field hygiene, yield losses have reduced significantly.

Tissue culture made it possible for farmers to have access to the following:

- large quantities of superior, clean, planting materials that are early maturing (12-16 months against 2-3 years for conventional material)
- heavier bunch weights (30-45 kg as to 10-15 kg from conventional material)
- higher annual yield per unit of land (40-60 tons/ha against 15-20 tons/ha with conventional material)

Benefits of TC for rice farmers in West Africa

Scientists have aimed to combine the ruggedness of African rice, *Oryza glaberrima* and the productivity of *Oryza sativa*. However, crossings failed as resulting offsprings were sterile. In the 1990s, the West Africa Rice Development Association (WARDA) used biotechnology to overcome the infertility problems.

After crossing, embryos were removed and grown on artificial media, a technique called "embryo-rescue." Since the resulting plants are frequently almost sterile, they were re-crossed with the sativa parent (backcrossing) whenever possible. Once the fertility of the progeny has improved after several cycles of backcrossing, anther culture was used to double the gene complement of the male sex cells (anthers) to produce true-breeding plants.

Their product, called 'New Rice for Africa' (NERICA), was made available in 1994 and since then, many lines have been generated. Most of the plants possessed the yield traits of the sativa as well as the adaptation traits of glaberrima.

Generally, NERICAs have the following characteristics:

- wide and droopy leaves
- longer and forked panicles or grain heads, which hold up to 400 grains
- more tillers with stronger stems
- higher yield than conventional rice
- mature 30 to 50 days earlier than current varieties
- taller than most rice varieties, resist pests, and tolerate drought better
- grow well on infertile and acidic soils
- have more body-building proteins than their parents

A study conducted between 2005 to 2011 estimates that 800,000 hectares of all Sub-Saharan Africa rice farms are planted to NERICA varieties.

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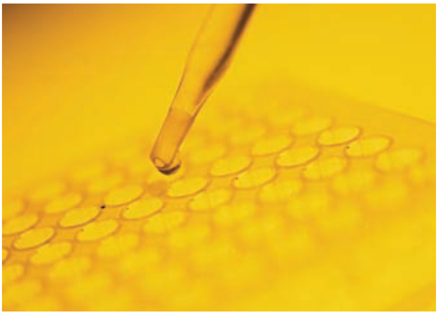
Plant Disease Diagnostics

Agricultural crops are threatened by various diseases and pests which can damage crops, lower harvest quality or even destroy entire harvests. Almost half of the world's total harvest is destroyed by diseases and pests annually. Moreover, farmers often deal with several pests or diseases and new pesticide-resistant pathogens.

If diagnosed early and correctly, treatments can be developed against these pathogens and could minimize losses. The common method of diagnosis is visual examination, which is often only possible after crops have been damaged. Hence, farmers should identify an infection before it becomes visible.

Pathogen infection in plants causes a complex immune response, producing proteins involved in plant defense. Pathogens also produce proteins and toxins to help their infection before disease symptoms appear. These molecules are crucial in the development of plant diagnostic kits.

Advances in molecular biology, plant pathology, and biotechnology have made the diagnostic kits possible. These detect diseases early, either by sensing the presence of the pathogen's DNA or the proteins produced by either the pathogen or the plant during infection. These require minimal processing time and are more accurate in identifying pathogens. Although some require laboratory equipment and training, other procedures can be done on-site by a person with no training.



Diagnostic kits have been designed to detect diseases in crops such as rice, potatoes, papaya, tomatoes, and banana. Similar kits are also used for identifying genetically modified organisms (GMOs) in shipments of conventional crops.

DNA-based diagnostic kits

DNA diagnostic kits are based on the ability of single stranded nucleic acids to bind to other single stranded nucleic acids with a complementary sequence. The tool used in DNA diagnostic kits is the Polymerase Chain Reaction (PCR). There are 3 steps involved in PCR:

1. The DNA is unwound, and its strands are separated by high temperatures
2. As the temperature is lowered, short, single-stranded DNA sequences called primers are free to bind to the DNA strands at regions of homology
3. This allows the (Taq) polymerase enzyme to make a new copy of the molecule.

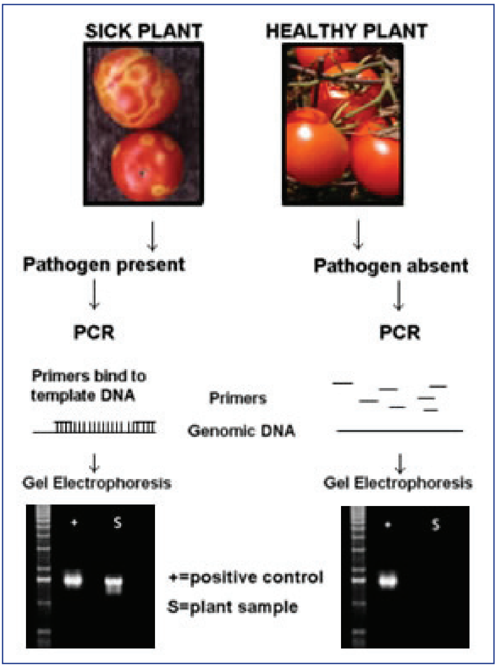


Figure 1. PCR-based diagnostic methods (Source: Alberts, et. al., 1994)

This cycle is repeated 30-40 times, yielding millions of identical copies of the segment. The primers in PCR diagnostic kits are very specific for the genes of a pathogen, and DNA amplification will occur only in diseased plants.

Several PCR-based methods (Figure 1) have successfully been adapted for plant pathogen detection such as the real-time PCR (RT PCR).

It follows the principle of PCR but quantifies amplified DNA using fluorescent dyes as it accumulates after each cycle. It offers several advantages over normal PCR, including reduced risk of sample contamination, real time data and simultaneous testing for multiple pathogens.

DNA microarrays are also of great use for simultaneous pathogen detection since plants are often infected with several pathogens at once, sometimes causing a disease complex. Microarrays consist of pathogen-specific DNA sequences immobilized onto a solid surface. Sample DNA is amplified by PCR, labeled with fluorescent dyes, and then hybridized to the array.

PCR-based diagnostics are sensitive enough to detect small amounts of DNA. PCR can also help farmers detect pathogens with long periods between infection and symptom development. Moreover, it can quantify pathogen biomass. PCR kits have been developed for black Sigatoka disease in bananas, *Phytophthora* infestations in potatoes, and *Fusarium* infection in cotton. However, PCR-based detection is expensive and requires expensive equipment.

Protein-based diagnostic kits

The first step in a defense response reaction is the recognition of an invader by the host's immune system. This recognition is due to the ability of host proteins, called antibodies, to recognize and bind proteins that are unique to a pathogen, called antigens, and trigger an immune reaction (Figure 2).

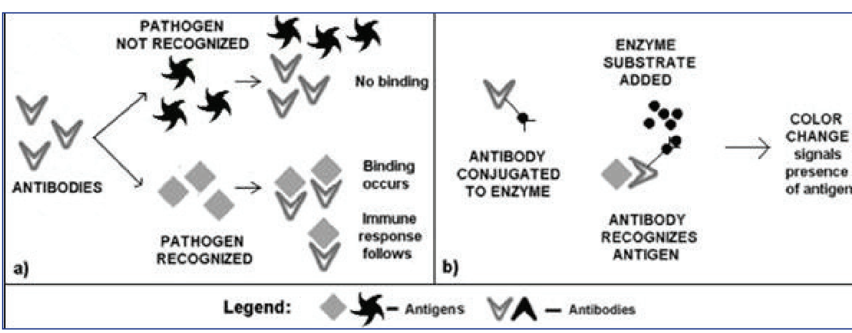


Figure 2. Antibody-antigen interaction (Source: Alberts, et. al., 1994)

Protein-based diagnostic kits for plant diseases contain a primary antibody that can recognize a protein from either the pathogen or the infected plant. It also contains a secondary antibody which is joined to an enzyme. This enzyme will catalyze a chemical reaction that causes a color change when the primary antibody is bound to an antigen signaling the presence of the pathogen.

The enzyme-linked immunosorbent assay (ELISA) method uses this detection system, and is the basis of some protein-based diagnostic kits. ELISA kits are very easy to use, takes only a few minutes to do, and does not require special laboratory equipment or training. There are already numerous ELISA test kits available on the market, including diagnostic kits for root crops, ornamentals, fruits, grains, and vegetables.

One of the first ELISA kits for diagnosing plant diseases was from the International Potato Center (CIP). It can detect the presence of all races, biovars, and serotypes of *Ralstonia solanacearum*, the causal agent of bacterial wilt or brown rot in potato. They also developed a kit that detects the presence of sweet potato viruses.

Conclusion

With even more advances in molecular biology and immunology, scientists and farmers alike can improve plant disease diagnosis. Development of better diagnostic kits for important crops is already underway. Diagnostic kits may be expensive but gains from it are definitely worth it. Their development should be made a priority in developing countries.

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Microbial Fermentation

For many years, man has worked to improve agricultural productivity by using soil microbes. These microbes can be cultivated on a large scale and made to assist plant growth. This process is known as microbial fermentation.

Microbes function as both providers and defenders, either by converting important macromolecules into forms usable by plants as biofertilizers or control weeds, pests, and diseases as bioherbicides and bioinsecticides.

Biofertilizers

Plants have a limited ability to extract phosphate and nitrogen from the environment and need microbes to absorb them. These microbes serve as biofertilizers in “nutrient recycling” and help plants gather energy sources in exchange for food in the form of by-products. This helps plants develop bigger root systems.

Colonies of *Penicillium bilaii* growing on a culture plate. (Source: http://www.dehs.umn.edu/iaq/fungus/penicillium/bilaii.html)

The fungus *Penicillium bilaii* produces an organic acid that converts phosphates into forms useful to plants. A biofertilizer from this organism is applied either by coating seeds with the fungus or applying it directly into the ground.

In legumes, the bacterium *Rhizobium* lives in nodules found in roots. These nodules can take nitrogen from the air and turn it into its available form and transfer the nutrient directly into the plant.

Biofertilizers have been found to:

- increase crop yield
- replace chemical nitrogen and phosphorus
- stimulate plant growth
- activate the soil biologically
- restore natural soil fertility
- protect against drought and some soil borne diseases

Bioinsecticides

Fermentation methods have also developed bioinsecticides which were based on the insecticidal proteins of bacteria, fungi, and viruses.

Bioinsecticides do not persist long in the environment, have short shelf lives and are effective even in small quantities. They are also safe to humans and animals and affect only a single species of insect.

However, bioinsecticides work slowly and their efficacy can depend on the timing of application. Since most bioinsecticide agents are living organisms, their success is also affected by environmental factors and other microbial competitors present in the environment.

Bacteria-based bioinsecticides

A widely used bioinsecticide, the bacterium *Bacillus thuringiensis*, or Bt, produces a protein poisonous to insects. After ingestion, the toxin creates ulcers in the insect's stomach causing the insect to die. However, Bt is very selective and affects only specific species of insect pests and does not harm humans, birds, fish, or other beneficial insects.

Fungi-based bioinsecticides

Fermentation technology is also used to mass produce fungi-based bioinsecticides. Their spores are harvested, packaged and applied to insect-infested fields. These spores use enzymes to break through the surface of the insects' bodies and, once inside, begin to grow and cause death.

One bioinsecticide, Bb, is based on the action of *Beauveria bassiana*, a fungus found worldwide in soils and plants. These Bb bioinsecticides have many advantages.

Beauveria bassiana fungus

The fungus does not grow in warm-blooded organisms, does not harm plants, and does not survive long in bodies of water. Its spores can also withstand harsh conditions and is inactivated by ultraviolet rays.

Virus-based bioinsecticides

An example of a virus-based bioinsecticide is the *Baculovirus*. It affects insect pests like corn borers, potato beetles, flea beetles, and aphids. One particular strain is being used to control Bertha army worms, which attack canola, flax, and vegetable crops.

Bioherbicides

Weeds are a constant problem for farmers and if left uncontrolled, can reduce crop yields significantly. Farmers fight weeds with tillage, hand weeding, synthetic herbicides, or a combination of all techniques.

The use of bioherbicides is another way of controlling weeds without the hazards of synthetic herbicides. Bioherbicides are made up of microorganisms and certain insects that can target specific weeds. The microbes possess invasive genes that can attack the defense genes of the weed and kill it.

Some bioherbicides also contain pathogens with genes that can cause fatal diseases to a specific weed only. This specificity of the microbes makes such bioherbicides very useful. Bioherbicides can also survive in the environment long enough for the next growing season and are cheaper than synthetic pesticides.

Bioherbicides and Striga

Striga flowers bloom just as the weed invades a cereal crop field. (IPM CRSP)

Sub-saharan Africa is home to sorghum and corn, as well as *Striga*, a weed that can wipe out important cereals, lower crop yields and increase the cost of production. Using bioherbicides together with genetic modification of certain cereals, scientists have lowered *Striga* parasitism and have increased corn and sorghum harvests.

For instance, the sorghum seeds can be inoculated with the fungus

Fusarium through a coating of Arabic gum. The preparation for this takes up to 14 days and is conducted by village women.

Another approach against *Striga* is the new hybrid maize Ua Kayongo, whose seeds are coated with Strigaway herbicide. It has Imazapyr resistance (IR-maize), which is based on a naturally-occurring herbicide resistance in maize and which was incorporated into Kenyan maize varieties by African plant breeders at International Maize and Wheat Improvement Center (CIMMYT) and the Kenya Agricultural Research Institute (KARI).

Conclusion

Microorganisms can either work symbiotically with plants to help in plant nutrition or they can work alone, or with other species, in battling weeds and pests. Although microorganisms are often labeled dangerous, they can be crucial in saving crops, increasing yields, and protecting soils.

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Genetic Engineering and GM Crops

Genetic engineering has developed rapidly in the last 30 years due to greater understanding of DNA and genes. Genetic engineering is the process in which the genetic makeup of an organism is altered using "recombinant DNA technology." This involves the use of laboratory tools to edit DNA.

Conventional plant breeding does not guarantee a particular gene combination will be obtained from crosses as undesirable genes can be transferred along with good genes or as a good gene is gained, another is lost. This random assortment of parents' genes in the offspring limits improvements that can be done.

Genetic engineering allows the direct transfer of genes of interest, between close or distant organisms to obtain the desired trait. However, not all genetic engineering techniques involve inserting DNA from other organisms as plants can also be modified by removing or switching off their own genes. Figure 1 shows a comparison of conventional breeding and genetic engineering.

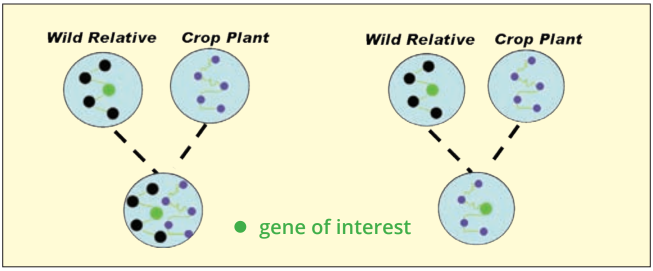

Figure 1. Conventional breeding (left) vs. genetic engineering (right)

Conventional Breeding

- Limited to the same or very closely related species
- Little or no guarantee of any particular gene combination from crosses done
- Undesirable genes can also be transferred
- Takes a long time to achieve desired results

Genetic Engineering

- Allows direct transfer of genes, between close or distant relatives
- Improvement can be achieved in a shorter time
- Allows plants to be modified by removing or switching off genes

Nature's own genetic engineer

The "sharing" of DNA among organisms is a natural phenomenon happening for thousands of years. *Agrobacterium tumefaciens*, a soil bacterium called the 'nature's own genetic engineer', is capable of genetically engineering plants. It causes crown gall disease, where large swellings (galls) occur at the crown of plants. The bacterium transfers part of its DNA to the plant which integrates into the plant's genome, causing the production of galls.

Application of genetic engineering in crop production

Genetic engineering techniques are used only if the trait to be introduced is not present in the germplasm of the crop, is very difficult to improve by conventional breeding methods or will take a very long time to introduce and/or improve in the crop by conventional methods. Crops developed through genetic engineering are called transgenic crops or genetically modified (GM) crops.

Modern plant breeding is a multi-disciplinary process that uses elements of conventional breeding, bioinformatics, molecular genetics, molecular biology, and genetic engineering.

Development of transgenic crops

There are five major steps in the development of a genetically engineered crop. But for every step, it is very important to know the mechanisms of action, regulation of gene expression, and safety of the gene and the gene product to be utilized. Before a genetically engineered crop is approved for commercial use, it has to pass rigorous safety and risk assessment procedures.

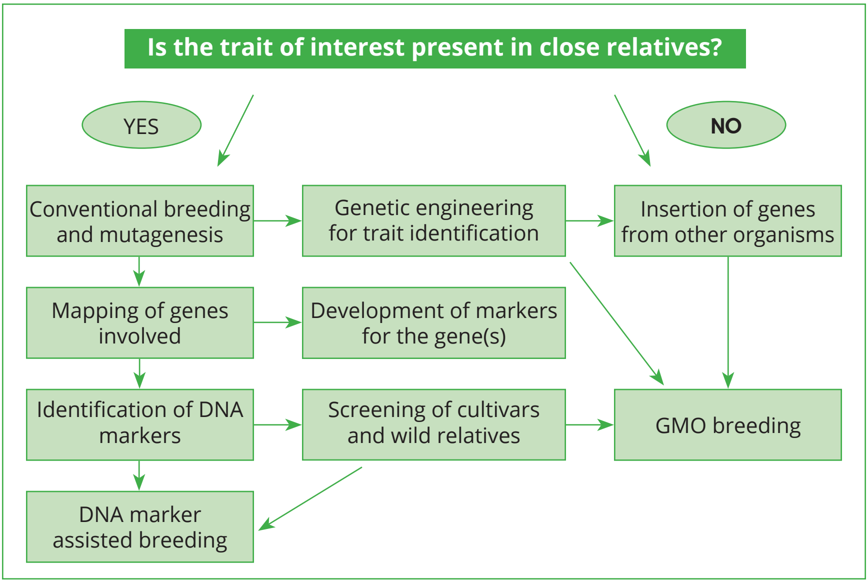

Figure 2. Integration of conventional and modern biotechnology methods in crop breeding (Source: DANIDA, 2002)

The first step is the extraction of DNA from the organism with the trait of interest. The second step is gene cloning, which isolates the gene of interest and clones it. Once cloned, the gene is designed so that it can be controlled and expressed inside the host plant. The modified gene will then be mass-produced in a host cell. Once ready, it can then be introduced into the cells of the plant being modified through *transformation*.

Common methods used to introduce the gene into plant cells include biolistic transformation (using a gene gun) or *Agrobacterium*-mediated transformation. Once the inserted gene is stable, inherited, and expressed in following generations, the plant is considered *transgenic*. Backcross breeding is the final step in the process, where the transgenic crop is bred to obtain high quality plants that express the inserted gene.

It may take up to 15 years, depending on the gene, crop and regulatory approval, before a transgenic hybrid is ready for commercial use.

Commercially available crops improved through genetic engineering

There has been a consistent increase in the global area planted to transgenic crops from 1996 to 2014. About 181.5 million hectares was planted in 2014 to transgenic crops. Transgenic crops with stacked traits, such as herbicide tolerant and insect resistant maize and cotton, are also available commercially.

New and future initiatives in crop genetic engineering

A number of products in the pipeline will soon make more direct contributions to food quality, environmental benefits and pharmaceutical production. Examples of these products include nutritionally-enhanced crops as well as abiotic stress resistant crops.

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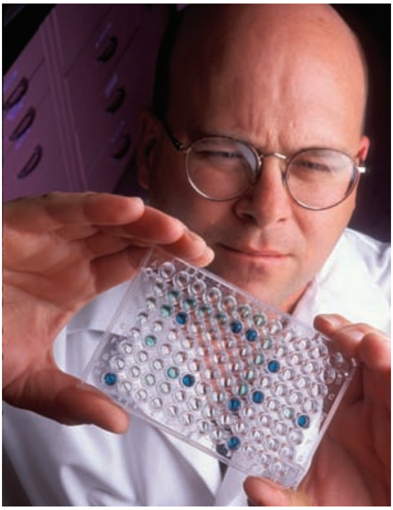
Molecular Breeding and Marker-Assisted Selection

Development of new crop varieties can take almost 25 years. However, biotechnology has considerably shortened the time by 7-10 years for new varieties to be brought to market. One of the tools that make it easier and faster to select plant traits is marker-assisted selection (MAS).

Molecular shortcut



The differences that distinguish one plant from another are encoded in the plant's genetic material, the DNA. These are packaged in chromosome pairs, one coming from each parent. The genes, which control characteristics, are located on specific segments of each chromosome. Together, all of the genes make up the genome.

Some traits may be controlled by only one gene. Others, however, like crop yield or starch content, may be influenced by several genes. Traditionally, plant breeders select plants based on visible or measurable traits, called the phenotype. However, this can be difficult, slow, and costly.

Plant breeders now use marker-assisted selection (MAS). To identify specific genes, scientists use molecular or genetic markers. Markers are sequences of nucleic acid which makes up a segment of DNA. Markers are located near the DNA sequence of the desired gene and are passed from generation to generation together with the desired gene. This is called genetic linkage. The presence of the marker also indicates the presence of the desired gene.

As scientists learn the locations of markers on a chromosome and their distance to genes, they create a genetic linkage map. This would show locations of markers and genes, and their distance from other genes.

Using detailed genetic maps, researchers are able to determine if a plant has the desired gene using just a piece of plant tissue from seedlings. If a plant doesn't have the desired gene, they are discarded until they only have plants with the gene.

However, molecular breeding through MAS is limited in scope compared to genetic engineering or modification because:

1. it works only for traits already present in a crop;
2. it cannot be used effectively to breed crops with long life cycles and;
3. it cannot be used effectively with crops that are propagated through cloning.

Molecular markers

Several marker systems have been developed and are applied to various crop species. These are the Restriction Fragment Length Polymorphisms (RFLPs), Random Amplification of Polymorphic DNAs (RAPDs), Sequence Tagged Sites (STS), Amplified Fragment Length Polymorphisms (AFLPs), Simple Sequence Repeats (SSRs) or microsatellites, and Single Nucleotide Polymorphism (SNPs). The advantages and disadvantages of these marker systems are shown in Table 1.

Table 1. Comparison of most commonly used marker systems (adopted from Korzun, 2003)

Feature	RFLPs	RAPDs	AFLPs	SSRs	SNPs
DNA required (ug)	10	0.02	0.5-1.0	0.05	0.05
DNA quality	High	High	Moderate	Moderate	High
PCR-based	No	Yes	Yes	Yes	Yes
Number of polymorph loci analyzed	1.0-3.0	1.5-50	20-100	1.0-3.0	1.0
Ease of use	Not easy	Easy	Easy	Easy	Easy
Amenable to automation	Low	Moderate	Moderate	High	High
Reproducibility	High	Unreliable	High	High	High
Development cost	Low	Low	Moderate	High	High
Cost per analysis	High	Low	Moderate	Low	Low

These techniques have been used to check differences in DNA sequences in and among species. They also allow the creation of new sources of variation by introducing desirable traits from wild varieties. While RFLP markers have been the basis for most genetic work in crops, AFLPs and SSRs are currently the most popular techniques used due to ease in detection and automation. The adoption of the new marker system, SNPs, is now highly preferred, with the increasing amount of sequence information, and the determination of gene function due to genomic research.

Applications of molecular markers for crop genetic studies

The main uses of these molecular markers in crop genetic studies are as follows:

- Assessment of genetic variability and characterization of germplasm
- Identification and fingerprinting of genotypes
- Estimation of genetic distances between population, inbreeds, and breeding materials
- Detection of monogenic and quantitative trait loci (QTL)
- Marker-assisted selection
- Identification of sequences of useful candidate genes

MAS for pathogen resistance in tomato

One of the major problems in tomato cultivation are severe harvest losses caused by several pathogens, including viruses, bacteria, fungi, and nematodes. Although conventional breeding has had a significant impact on improving tomato resistance, the long duration of breeding makes it difficult to cope with new virulent pathogens.

Molecular markers are now being used for breeding tomato. More than 40 genes that confer resistance to tomato pathogens have been mapped, cloned, and/or sequenced. These maps have allowed for "pyramiding" resistance genes in tomato through MAS, where several resistance genes can be engineered into one genotype. Currently, tomato breeding through MAS has resulted in varieties with resistance or tolerance to one or more specific pathogens.

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Gene Switching and GURTs: What, How, and Why?

Genetic Use Restriction Technologies (GURTs) is an ongoing topic of discussion under the Convention on Biological Diversity (CBD). The current focus surrounding this topic concerns whether and how GURTs may impact indigenous people, local communities and small-holder farmers. Indeed, in the most recent debate on this topic held in February 2005, the representatives of the indigenous people and local communities requested clear and objective information on GURTs so that they could understand the issues and better participate in the discussion.

The following narrative seeks to respond to that request by explaining what gene switching and GURTs are, how they work, and why public and private sector scientists, as well as governments, are pursuing further research and development in this area.

What is gene switching and how does it work?

Biotechnology-based gene switching is the use of genetic engineering to control specific genes in plants to achieve desired results. These targeted genes are controlled through “switch mechanisms” which activate, deactivate, or adjust upward or downward gene functions.

It is also described as controlling the “expression” of genes. Gene switching mechanisms are established either in response to an external trigger, to activate the genes at critical times, or in particular locations in the plant. It should be called “biotechnology-based gene switching” as gene switching occurs naturally and without human intervention.

Researches have focused on gene switching applications to control genes related to specific plant traits. In these cases, all the other genes in the plant continue to function normally. Seed from these plants could be saved by farmers and planted the next year, however, it often results in a crop without the special trait.

The biotechnology-based gene switching applications also include controlling genes for reproduction or seed germination. It controls plant reproduction either by limiting pollen production or by producing non-viable seeds. Examples of this are seedless grapes and watermelons.

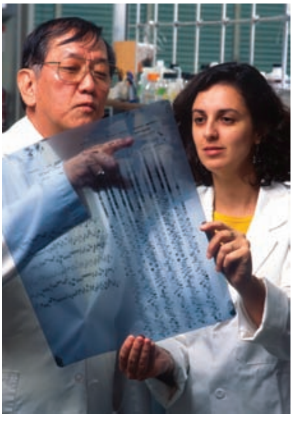
Why are scientists exploring gene switching?

Researchers are exploring the use of gene switching to allow plants to express a gene only when it is needed. For instance, a drought tolerant plant will produce the gene for drought tolerance only when drought occurs.

Other examples include the following:

- Sentinel plants that would notify farmers when there is a nutrient deficiency or a pest infestation in their fields
- Targeted release of Bt or other pest protection mechanisms within a plant, helping to further reduce the potential development of pest resistance
- The development of sterile progeny, further contributing to environmental risk management processes in centers of origin and other sensitive environments

How are gene switching technologies regulated?



National biosafety frameworks regulate viable products of genetic engineering, including any plants and organisms that may be created through gene switching technologies, on a case-by-case basis through scientific risk assessment. Accordingly, any unique attributes of products of gene switching, including those that result in sterile seed, automatically are considered in risk assessment and decision-making. Based on the Cartagena Protocol on Biosafety, gene switching products are continued to be regulated under the same biosafety system as any other biotechnology application.

What is the purpose of producing sterile seeds?

GURTs produce genetically engineered plants that cannot develop viable seeds, thus preventing unintended introduction to the environment. A number of government bodies have recognized this potential biosafety benefit of GURTs, and funds have been allocated to support additional research.

Companies produce sterile seeds to protect its technology and investment through preventing unauthorized saving of seeds for subsequent years. The farmer who purchases this seed will know that he will not be able to save seeds because these will be labelled by manufacturers with information on restrictions related to patents. However, GURTs products may cost more than conventional seeds. Some farmers still choose to buy these seeds because of particular benefits they offer.

Conclusion

Biotechnology-based gene switching in plants describes a wide range of mechanisms to control gene expression for purposes beneficial to human beings and the environment. These technologies hold promise to more efficient and effective traits in plants. The technology also offers an additional layer of biosafety protection as well as serving to protect research and development investments.

All genetically modified organisms created through biotechnology-based gene switching can and should be reviewed and assessed on a case by case basis, under scientifically sound regulatory frameworks, in line with existing CBD guidance.

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Marker-Free GM Plants

Selectable marker genes are vital in developing genetically modified crops since pinpointing cells with foreign DNA among non-transformed cells is very hard. To find transgenic cells, marker genes are co-introduced with the transgene. These genes usually confer resistance to antibiotics and herbicides.

The presence of marker genes in commercialized transgenics has caused concern about the safety of GM crops. Herbicide resistance genes could be transferred to other crops while antibiotic resistance genes could transfer to microorganisms, resulting in resistant pathogens. The difficulty of proving that marker genes are harmless has limited the acceptance of biotechnology.

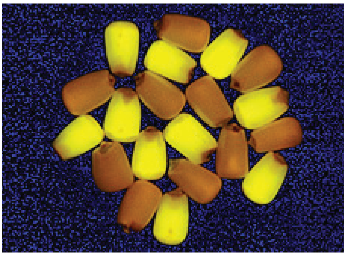
Researchers have been trying to develop marker-free transformation methods and selectable marker elimination strategies. This would minimize public concerns and reduce costs for developing GM products and hasten commercial release.

There are several ways to avoid or eliminate selectable marker genes such as site-specific recombination, transposition and homologous recombination.

Alternatives to antibiotic/herbicide resistance markers

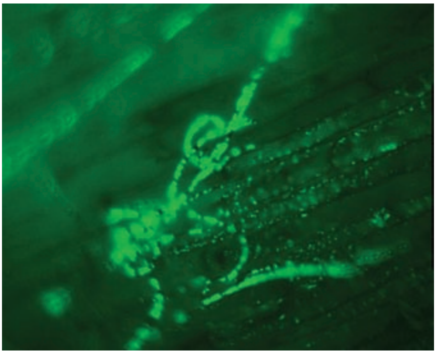
Scientists identified selectable marker genes that are dependent on non-toxic substances and could be substrates for transformed cells. These markers would only suppress the growth of non-transformed cells.

For instance, events can be selected using markers that enable them to use a specific food. An example is the use of phosphomannose isomerase gene (pmi). Transformed cells can be identified since they can use mannose as food.



Fluorescent protein is introduced into corn as marker for different tissues, to study nitrogen use and grain development. The light kernels express the fluorescent protein in the endosperm layer.

Genes that allow plants to survive in media with amino acid analogs have also been used. The use of alternative markers eliminates environmental concerns. However, they require a more rigorous risk assessment.



A microscopic image of the beneficial fungus *Metarhizium anisopliae* showing a fluorescent green protein as it grows on the surface of a young sugar beet root.

Researchers have also used markers to make transgenic plants visually recognizable. The green florescent protein (GFP) gene from jellyfish makes transformed cells appear green when exposed to ultraviolet light. Reporter genes, like the firefly protein luciferase, have also been used as visible markers. However, the transformed and non-transformed cells must be manually separated in this approach.

Conventional genetics lends a hand: Co-transformation

One of the simplest marker removal strategies is the co-transformation approach. The principle of this is to integrate the transgene and the marker gene into unlinked locations in the genome and which may segregate in the next generation to yield progenies with the transgene without the markers.

Three approaches are used for co-transformation: introducing two *Agrobacterium* strains, one with the marker and another with the transgene; using one bacterial strain with two vectors, each with one gene; and using a bacterial strain with one vector containing the two genes at separate sites.

However, since it relies on segregation during sexual reproduction, it cannot be used for vegetatively propagated plants. Selection of the progenies carrying only the target gene is also laborious.

Molecular cut and paste: Site specific recombination

Microbial site specific recombinases have also been used to eliminate markers from GM plants. These act as scissors capable of cleaving DNA at specific sites. They can also act as glue, ligating the cleaved DNA fragments at another target sequence. The gene encoding these enzymes is introduced along with the marker gene and the transgene. Once transformed cells are selected, the recombinase gene is activated by external stimulus. The recombinases then cut out the marker genes and the genes for the enzymes themselves.

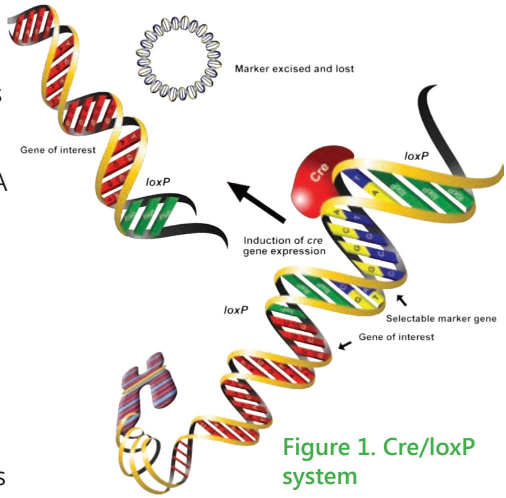


Figure 1. Cre/loxP system

The most widely used site-specific recombination system is the *Cre/loxP* system (Figure 1). The Cre recombinase catalyzes a reaction between two *loxP* sequences and results in excision of the DNA fragment between them. The Cre recombinase gene can be introduced into transgenic plants by re-transformation, breeding or inducible autoexcision.

Jumping along chromosomes: Marker deletion via transposons

The process that enables certain genes to 'jump' at a certain position on the genome can also be used to generate marker-free plants. The approach is similar to site-specific recombination only that transposons or jumping genes are used. Transposons contain a gene for a special enzyme, which recognizes signals in the DNA. The enzyme cuts the DNA fragment beside these signals and integrates them randomly in the genome. The most notable transposons are the Ac/Ds family, the enzyme being the Ac (activator) transposase and the Ds (dissociator) sequences the tag signals.

The gene of interest or the marker gene can be placed within the 'jumping' sequence, in such a way that the two genes can be separated from each other upon the activation of transposase. Although effective, its efficiency is poor due to the low incidence of occurrence. It can also be time-consuming since breeding is required to separate the transgene and the marker.

Future prospects

Numerous approaches to eliminate antibiotic and herbicide markers have been developed over the years and further improvements are still underway. Scientists are also searching for ways to hasten the selection of marker-less progenies. Novel marker elimination strategies based on gene targeting and homologous recombination have been reported. With these, the concern about the spread of antibiotic and herbicide resistance genes in the environment might become irrelevant in the future.

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RNAi for Crop Improvement

RNA interference (RNAi) is a method of blocking gene function by inserting short sequences of ribonucleic acid (RNA) that match part of the target gene's sequence and thus producing no proteins. RNAi has emerged as the method for researchers studying the structure and function of genes.

RNAi has a huge potential as a powerful approach in targeted and personalized medicine as well as in agriculture. RNAi has provided a way to control pests and diseases, introduce traits and increase crop yield. Scientists have developed novel crops such as nicotine-free tobacco, and nutrient-fortified maize through RNAi.

Discovery of RNAi

Scientists believed they could produce any gene product simply by introducing foreign genes in plants.[1] In a previous study, biologists introduced multiple copies of the gene for purple petunia flowers to create deep purple flowers. This instead resulted in plants with white or variegated flowers. The transgenes were silenced as well as the plant's 'purple-flower' gene.[2,3]

The mechanism that caused this effect was discovered by Andrew Fire and Craig Mello when their injection of double stranded ribonucleic acids (dsRNA) into the worm *Caenorhabditis elegans* triggered silencing of genes with sequences identical to that of the dsRNA.[4] They called the phenomenon RNA interference. Fire and Mello were awarded the 2006 Nobel price for Physiology or Medicine for their discovery.

In addition to its roles in regulating gene expression, RNAi is used as an immune response to infection[5] and as a natural defense mechanism against molecular parasites such as jumping genes and viral genetic elements that affect genome stability.[6] Specific types of bacteria have also been shown to trigger the RNAi pathway in plants.

How RNAi works

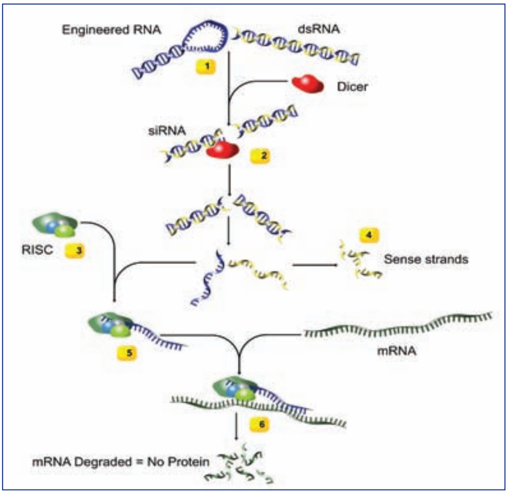

Figure 1. Mechanism of RNAi

1. The entry of any long double stranded RNA triggers the RNAi pathway of cells. This results in the recruitment of the enzyme Dicer (Figure 1).
2. Dicer cleaves the dsRNA into short, 20-25 base pair-long fragments, called small interfering RNA (siRNA).
3. An RNA-induced silencing complex (RISC) then separates the siRNA strands into two: sense or antisense strand. The sense strands, or those with exactly the same sequence as the target gene, are degraded.
4. The antisense strands are incorporated to the RISC and are used as guide to target messenger RNAs (mRNA).
5. mRNA, which codes for amino acids, are cleaved by RISC. The activated RISC can repeatedly participate in mRNA degradation, inhibiting protein synthesis.

Disease and pathogen resistance

Gene silencing was first used to develop virus-resistant varieties. This was first demonstrated in Potato virus Y- resistant plants with RNA transcripts of a viral proteinase gene.[7,8] Plants can also be modified to produce dsRNAs that silence genes in crop pests. This was used to develop resistance to root-knot nematode[9], corn rootworm[10] and cotton bollworm.[11]

Male sterility

RNAi has also been used to develop male sterility, which is vital in the hybrid seed industry. Genes expressed solely in tissues involved in pollen production are targeted through RNAi. Scientists developed male sterile tobacco by inhibiting the expression of *TA29*, a gene necessary for pollen development.[12]

Plant functional genomics

RNAi offers specificity and efficacy in silencing members of a gene or multiple gene family to characterize a gene function. The expression of dsRNAs with inducible promoters can also control the extent and timing of gene silencing, resulting in silenced genes at a certain growth stage or plant organ.[13,14] There are several ways of activating the RNAi pathway in plants such as the use of hairpin RNA-expressing vectors, particle bombardment, *Agrobacterium*-mediated transformation and virus-induced gene silencing (VIGS).[15]

Engineering plant metabolic pathways

RNAi has been used to modify plant metabolic pathways to enhance nutrient content and reduced toxin production (Table 1). The technique uses heritable and stable RNAi phenotypes in plants.

Prospects for RNAi

RNAi presents the possibility of targeting multiple genes for silencing using a thoroughly-designed single transformation construct. It can also provide resistance against a wide range of pathogens.[9] Studies have also used it in plant stress adaptation.

Table 1. Examples of novel plant traits engineered through RNAi

Trait	Target Gene	Host	Application
Enhanced nutrient content	Lyc	Tomato	Increased concentration of lycopene (carotenoid antioxidant)
	DET1	Tomato	Higher flavonoid and b-carotene contents
	SBEII	Wheat, Sweet potato, Maize	Increased levels of amylose for glycemic management and digestive health
	FAD2	Canola, Peanut, Cotton	Increased oleic acid content
	SAD1	Cotton	Increased stearic acid content
	ZLKR/SDH	Maize	Lysine-fortified maize
Reduced alkaloid production	CaMXMT1	Coffee	Decaffeinated coffee
	COR	Opium poppy	Production of non-narcotic alkaloid, instead of morphine
	CYP82E4	Tobacco	Reduced levels of the carcinogen nornicotine in cured leaves
Heavy metal accumulation	ACR2	Arabidopsis	Arsenic hyperaccumulation for phytoremediation
Reduced polyphenol production	s-cadinene synthase gene	Cotton	Lower gossypol levels in cottonseeds, for safe consumption

Ethylene sensitivity	LeETR4	Tomato	Early ripening tomatoes
	ACC oxidase gene	Tomato	Longer shelf life because of slow ripening
Reduced allergenicity	Arah2	Peanut	Allergen-free peanuts
	Lolp1, Lolp2	Ryegrass	Hypo-allergenic ryegrass
Reduced production of lachrymatory factor synthase	lachrymatory factor synthase gene	Onion	"Tearless" onion

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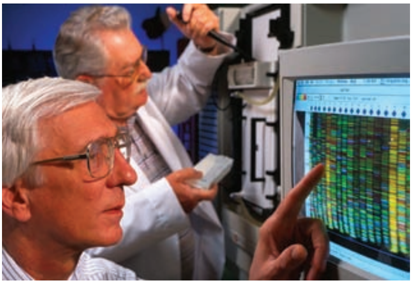
Bioinformatics for Plant Biotechnology

Scientists worldwide have already published complete genomes, and there are more genome projects currently ongoing. These studies have helped to understand plant evolution, and are used to improve crops. With all this genetic information, scientists need databases and related tools to manage, analyze and use the information. This is the role of bioinformatics.

What is bioinformatics?

Bioinformatics is a field of science that combines computers, mathematical algorithms, and statistics with concepts in the life sciences to analyze various genomes. Examples of these include those of maize and citrus species.

What data does bioinformatics deal with?



Bioinformatics deals with the following data:

1. DNA, RNA, and protein sequences — sequence of nucleotides in DNA or RNA, and sequence of amino acids in a protein
2. Molecular structures — Higher molecular structure; these data are obtained by combining thermodynamic data and computer modeling with data from laboratory techniques
3. Expression data — data on when and where genes are expressed as well as overall gene expression in certain cells or in specific environmental conditions
4. Bibliographic data — research projects and genome sequencing programs organized in public online databases

What can bioinformatics do with this data?

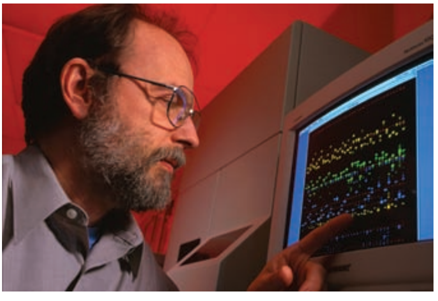
Understanding biological sequences and structures begins with data management. It is the most fundamental task of bioinformatics. This is done by transferring information into a database.

A database is a collection of information stored systematically. In bioinformatics, this includes DNA, RNA, or protein sequences. Sequences can be sorted according to their function, species of origin, or articles where they were first reported. The database may also contain journal articles and abstracts.

After data management, bioinformaticians can now mine, retrieve, and use the data. This is done through computer programs that search databases and retrieve needed information.

How can bioinformatics improve plant biotechnology?

It can aid scientists in basic research



Complete sequence of a plant's genome can lead to future studies for that plant. For instance, the USDA-Agricultural Research Service (USDA-ARS) analyzed gene expression patterns in soybean and barley to determine the function of genes involved in stress resistance in plants.

It can be used to design better plants

If the genes for certain traits are known, scientists can design methods for improving crops. Bioinformatics helps scientists design plants with higher quality fruit or with tolerance to extreme environmental conditions.

Researchers from Australia's Queensland Agricultural Biotechnology Center, for example, looked at expressed sequence tags (EST) to determine genes involved in papaya ripening and design ways to produce better papayas.

It can be used to harness genetic diversity

Wild relatives of plants are potential sources of desirable genes. By

knowing closely related plants, scientists can identify sexually compatible species with desirable characteristics. For instance, researchers at the Weizmann Institute in Israel studied the gene exchange between crops and their wild relatives and used it to incorporate desirable genes into modern crops.

It can be used to design new tools to study gene function
MicroRNAs (miRNAs), gene sequences that are vital to plant growth, target certain DNA sequences and keep certain genes inactive. These molecules are now used to silence gene families to study their function.

It can be used to test, analyze, and identify plants
With microarray profiles available online, scientists can learn about differences in gene expression in normal and stress conditions. If certain genes are highly expressed during stress conditions, they may be crucial to the plant's survival and may be used to improve other plants without that gene.

Scientists also perform protein or RNA profiling to compare GM plants to conventional plants. Studies have compared GM and conventional potato by analyzing their proteome and found no new proteins unique to GM lines.

Bioinformatics at your fingertips: The NCBI Online

The National Center for Biotechnology Information (NCBI) is an online resource and database for scientists, researchers, and the general public. It is full of tools that can aid interested parties in doing the following:

Search - NCBI has a search engine called the Basic Local Alignment Search Tool (BLAST). This is similar to others, except that the queries are nucleotide (BLASTn) or protein (BLASTp) sequences. Scientists can use BLAST to look for DNA or protein sequences. Search matches can then tell them vital information such as what their gene or protein is, its source organism, and other organisms with the same gene or protein sequence.

Research - ENTREZ is the search and retrieval system used at NCBI for its databases. It helps scientists find out how many genes or proteins of interest are publicly available, how many of such genes or proteins have been sequenced in a given organism, and what research has already been published in the field.

Add - Sequence "depositors" can add sequences to the database through an online tool.

Mine - NCBI has several bioinformatics tools designed to mine data from the database. For instance, Spidey can align RNA sequences to a genomic sequence and determine where the gene ends and other sequences begin.

The way forward

Bioinformatics not only provides information, but leads to more experiments. For instance, a previous study from Iowa State University investigated unique sequences in the oat genome. This allowed the researchers to find specific regions of DNA that would both identify oat types through PCR, as well as serve as markers in marker-assisted selection.

Conclusion

There are many tools in bioinformatics, each functions to suit the needs of those using them. Gene and protein databases are constantly updated with information that aid scientists all around the world. Bioinformatics benefits plant breeding and genetic engineering by allowing scientists to produce better crops for the future.

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Nanotechnology in Agriculture

Nanotechnology refers to controlling, building, and restructuring materials and devices on the scale of atoms and molecules.[1] A nanometer (nm) is one-billionth of a meter. At nano scales, the basic rules of chemistry and physics are not applicable.[2] An example of this technology is the carbon nanotube (Figure 1) discovered in 1991, which conducts electricity better than copper, is stronger than steel but significantly lighter.[3]

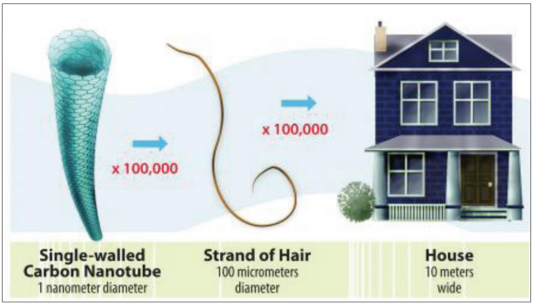


Figure 1. Carbon nanotube (Source: nano.gov/nanotech-101/what/nano-size)

Nanotechnology for crop biotechnology

Chemists have developed DNA crystals by producing synthetic DNA sequences that can self-assemble into three-dimensional triangle-like patterns. The DNA crystals have "sticky-ends" that can attach to another molecule in an organized fashion. When multiple helices are attached, it will form a three-dimensional crystal (Figure 2). This could be applied in improving important crops by organizing and linking carbohydrates, lipids, proteins and nucleic acids to these crystals.[4]

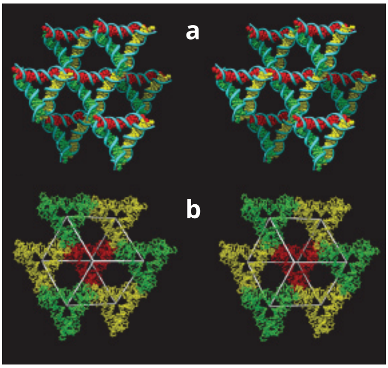
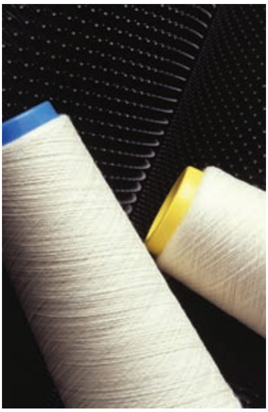


Figure 2. a. Stereographic image of the surrounding of a triangle; b. Stereographic image of the rhombohebral cavity (white lines) formed by the triangles. (Source: Nature)

Nanoparticles can serve as 'magic bullets', containing herbicides, chemicals, or genes, which target particular plant parts to release their content. Nanocapsules can enable effective penetration of herbicides, allowing slow and constant release of the substance.[5] Iowa State

University researchers used chemically-coated 3-nm mesoporous silica nanoparticles (MSN) as containers for genes incorporated into plants. The coating allows the plant to take the particle, where the genes are inserted. This technique has been used in tobacco and corn.[6]

Nanoparticles and recycling agricultural waste



Nanotechnology is also used to reduce waste in agriculture. In cotton production, cellulose are often discarded as waste or used for low-value products. Using nanotechnology, this cellulose is used to produce nanofibers that can act as a fertilizer or pesticide absorbent, which allows targeted application at a specific time and location.[7]

Nanotechnology is also used to enhance the performance of enzymes used in ethanol production. Scientists are also working on nanoengineered enzymes that allow simpler and cheaper production of ethanol.[8]

When burned, rice husk produces high-quality nanosilica which can be used in making other materials such as glass and concrete. Mass production of nanosilica through nanotechnology can alleviate the growing rice husk disposal concern.[9]

Delivery systems for pests, nutrients, and plant hormones

Nanosensors and nano-based smart delivery systems also help in the efficient use of agricultural natural resources through precision farming. Using nanomaterials and global positioning systems with satellite imaging of fields, farm managers could detect pests or stresses. Once detected, an automatic adjustment of

Center-pivot irrigation system used in precision farming pesticide applications or irrigation would occur. Nanosensors in the field can also detect plant viruses and soil nutrition. Nanoencapsulated slow-release fertilizers have also been used to save fertilizer.[10]

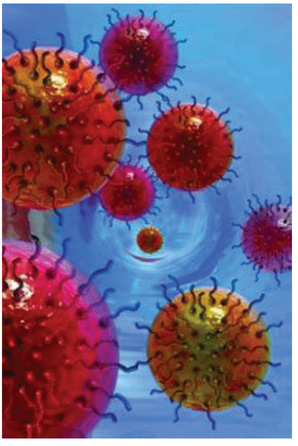
Nanobarcodes and nanoprocessing are also used to monitor the quality of agricultural produce. Cornell University researchers aimed to develop an on-site detector which can be used by non-trained people. They produced microscopic probes or nanobarcodes that could tag pathogens in a farm. Once tagged, these are easily detected using any fluorescent-based equipment.[11]

Nanotechnology also allowed scientists to study a plant's hormone regulation. Scientists at Purdue University developed a nanosensor that generates an electrical signal that measures auxin concentration at a particular part. This breakthrough helps scientists understand how plant roots adapt to the environment.[12]

The impact of nanotechnology

Nanotechnology is one of the possible solutions to problems in food and agriculture. Like biotechnology, safety issues and regulation are raised on nanotechnology. However, nanotechnology products have long been commercially available.[13]

Nobel laureate Richard Smalley presented the benefits of nanotechnology to the U.S. House Committee on Science in 1999. He emphasized that the impact of nanotechnology on health, wealth, and lives of the people will be at least equal to the combined influences of microelectronics, medical imaging, computer-aided engineering and man-made polymers developed in the 20th century.[3] To date, nanoscientists are developing techniques for atom-by-atom construction of objects that have potential applications not just in agriculture but also in medicine, electronics, information technology, and environmental monitoring and remediation, to name a few.[14]



Nanorust cleans arsenic from drinking water (Source: Rice University)

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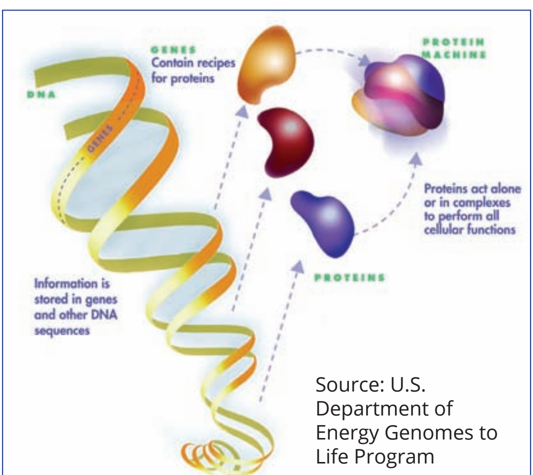
'Omics' Sciences: Genomics, Proteomics, and Metabolomics

Genomics

Genomics is the field of science that deals with the discovery and noting of all the sequences in the entire genome of a particular organism. The genome can be defined as the complete set of genes inside a cell. Therefore, genomics is the study of the genetic make-up of organisms.

Determining the genomic sequence, however, is only the beginning of genomics. Once this is done, the genomic sequence is used to study the function of the numerous genes (functional genomics), to compare the genes in one organism with those of another (comparative genomics), or to generate the 3-D structure of one or more proteins from each protein family, thus offering clues to their function (structural genomics).

In crop agriculture, the main purpose of the application of genomics is to gain a better understanding of the whole genome of plants. Agronomically important genes may be identified and targeted to produce more nutritious and safe food while at the same time preserving the environment.



Source: U.S. Department of Energy Genomes to Life Program

Figure 1. Genes, proteins, and molecular machines

Genomics is an entry point for looking at the other 'omics' sciences. The information in the genes of an organism, its genotype, is largely responsible for the final physical makeup of the organism, referred to as the "phenotype". However, the environment also has some influence on the phenotype.

DNA in the genome is only one aspect of the complex mechanism that keeps an organism running — so decoding the DNA is one step towards understanding the process. However, by itself, it does not specify everything that happens within the organism.

The basic flow of genetic information in a cell is as follows: the DNA is transcribed or copied into a form known as “RNA”. The complete set of RNA (also known as its transcriptome) is subject to some editing (cutting and pasting) to become messenger-RNA, which carries information to the ribosome, the protein factory of the cell, which then translates the message into protein.

Proteomics

Proteins are responsible for an endless number of tasks within the cell. The complete set of proteins in a cell can be referred to as its proteome and the study of protein structure and function and what every protein in the cell is doing, is known as proteomics. The proteome is highly dynamic and it changes from time to time in response to different environmental stimuli. The goal of proteomics is to understand how the structure and function of proteins allow them to do what they do, what they interact with, and how they contribute to life processes.

In an application of proteomics known as protein “expression profiling”, proteins are identified at a certain time in an organism as a result of the response to a stimulus. Proteomics can also be used to develop a protein-network map where interaction among proteins can be determined for a particular living system. Proteomics is also applied in mapping protein modifications to determine the difference between wild types and genetically modified organisms. It is also used to study protein-protein interactions involved in plant defense reactions.

A proteomics study was conducted in Iowa State University, where they examined the changes of protein in corn proteome during low temperatures. In another study, researchers analyzed the differences in the genome expressions of developing soybean under high temperature and identified the proteins expressed in response to diseases.

Metabolomics

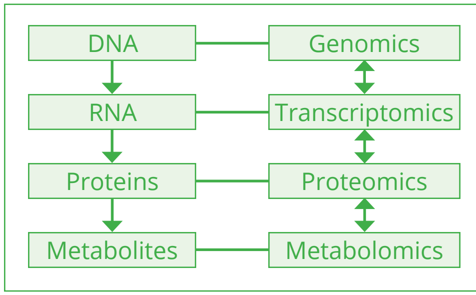


Figure 2. The taxonomy of 'omics' sciences

Metabolomics aims to determine a sample's metabolome, the complete set of low molecular weight compounds which are the substrates and by-products of enzymatic reactions and directly affects the phenotype of the cell, at a specified time under specific environmental conditions. While genomics and proteomics provide extensive information regarding the genotype, they convey limited information about phenotype. Low molecular weight compounds are the closest link to phenotype.

Metabolomics can be used to determine differences between the levels of thousands of molecules between a healthy and diseased plant. This can also be used to determine the nutritional difference between traditional and genetically modified crops, as well as in identifying plant defense metabolites.

Genomics provides an overview of the complete set of genetic instructions provided by the DNA, while transcriptomics looks into gene expression patterns. Proteomics studies dynamic protein products and their interactions, while metabolomics is also an intermediate step in understanding organism's entire metabolism.

The International Rice Genome Sequencing Project

This genomic research in rice is a collaborative effort of several public and private laboratories worldwide. This project completely sequenced the entire rice genome (12 rice chromosomes) and subsequently applied the knowledge to improve production.

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GMO Facts

Q&A about Genetically Modified Crops

Global agriculture finds itself engrossed in a heated debate over genetically modified (GM) crops. This debate, which features science, economics, politics, and even religion, is taking place almost everywhere. It is going on in research labs, corporate boardrooms, legislative chambers, newspaper editorial offices, religious institutions, schools, supermarkets, coffee shops, and even in private homes.

What is all the fuss about and why do people feel so strongly about this issue? This Q&A attempts to shed light on the controversy by addressing basic questions about GM crops.

What is a GM crop?

A GM or transgenic crop is a plant that has a novel combination of genetic material obtained through the use of modern biotechnology.

For example, a GM crop can contain a gene(s) that has been artificially inserted instead of the plant acquiring it through pollination.

The resulting plant is said to be "genetically modified" although in reality all crops have been "genetically modified" from their original wild state by domestication, selection, and controlled breeding over long periods of time.

Why make GM crops?

Traditionally, a plant breeder tries to exchange genes between two plants to produce offspring that have desired traits. This is done by transferring the male (pollen) of one plant to the female organ of another.

This cross breeding, however, is limited to exchanges between the same or very closely related species. It can also take a long time to achieve desired results and frequently, characteristics of interest do not exist in any related species.

GM technology enables plant breeders to bring together in one plant useful genes from a wide range of living sources, not just from within the crop species or from closely related plants. This powerful tool allows plant breeders to do faster what they have been doing for years — generate superior plant varieties — although it expands the possibilities beyond the limits imposed by conventional plant breeding.

Who produces GM crops?

Most of the research on GM crops has been carried out in developed countries, mainly in North America and Western Europe. However, many developing countries have also established the capacity for genetic engineering and have several products in the pipeline.

In developed countries, the life sciences companies have dominated the application of GM technology to agriculture. These include Bayer CropScience, BASF, Dow AgroSciences, DuPont/Pioneer, Limagrain, Monsanto, and Syngenta.

Where are GM crops currently grown?

In 1994, Calgene's delayed-ripening tomato (Flavr-Savr™) became the first GM food crop to be produced and consumed in an industrialized country. Since the recorded commercialization of GM crops in 1996 to date, several countries have contributed to 100-fold increase in the global area of biotech crops.

The countries growing biotech crops are: U.S., Brazil, Argentina, India, Canada, China, Paraguay, Pakistan, South Africa, Uruguay, Bolivia, Philippines, Australia, Burkina Faso, Myanmar, Mexico, Spain, Colombia, Sudan, Honduras, Chile, Portugal, Cuba, Czech Republic, Romania, Slovakia, Costa Rica, Bangladesh, and Vietnam.

What are the potential benefits of GM Crops?

In the developed world, there is clear evidence that the use of GM crops has resulted in significant benefits. These include:

- Higher crop yields
- Reduced farm costs
- Increased farm profit
- Improvement in health and the environment

These "first generation" crops have proven their ability to lower farm-level production costs. Now, research is focused on "second-generation" GM crops that will feature increased nutritional and/or industrial traits. These crops will have more direct benefits to consumers. Examples are:

- Rice enriched with iron, vitamin A and E, and lysine
- Potatoes with higher starch content, and inulin
- Edible vaccines in maize, banana, and potatoes
- Maize varieties with low phytic acid and increased essential amino acids
- Healthier oils from soybean and canola
- Allergen-free nuts

Are GM crops appropriate for developing countries?

While most of the debate over transgenic crops has taken place mainly in the developed nations in the North, the South stands to benefit from any technology that can increase food production, lower food prices, and improve food quality.

In countries where there is often not enough food to go around and where food prices directly affect the incomes of majority of the population, the potential benefits of GM crops cannot be ignored. It is true that nutritionally enhanced foods may not be a necessity in developed countries but they could play a key role in helping to alleviate malnutrition in developing countries.

Although the potential benefits of GM crops are large in developing countries, they would require some investments. Most developing countries lack the scientific capacity to assess the biosafety of GM crops, the economic expertise to evaluate their worth, the regulatory capacity to implement guidelines for safe deployment, and the legal systems to enforce and punish transgressions in law. Thus, several organizations are working to build local capacity to manage the acquisition, deployment, and monitoring of GM crops.

Conclusion

Despite the current uncertainty over GM crops, one thing remains clear. This technology, with its potential to create economically important crop varieties, is simply too valuable to ignore. There are, however, some valid concerns. If these issues are to be resolved, decisions must be based on credible, science-based information.

Finally, given the importance people place on the food they eat, policies regarding GM crops will have to be based on an open and honest debate involving a wide cross-section of society.

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Plant Products of Biotechnology

Biotechnology products have been in the market for some time now. These modified crops look like their conventional counterparts, but they possess special traits that make them better. These crops give benefits to both farmers and consumers. Farmers get higher crop yields and have increased flexibility in management practices while consumers have “healthier crops” (i.e., crops grown with fewer pesticides and/or with healthier nutritional characteristics).

Plant products of biotechnology (Table 1) approved for food use have been modified to contain traits such as insect resistance, disease resistance, herbicide tolerance, altered nutritional profile, and enhanced storage life.

Table 1. Examples of plant products of biotechnology

Crop	Trait(s)
Alfalfa	Herbicide tolerance (HT), altered lignin production
Apple	Non-browning
Bean	Viral disease resistance
Argentine canola	HT, modified oil/fatty acid, phytase production, pollination control system
Cotton	HT, insect resistance (IR)
Creeping bentgrass	HT
Eggplant	IR
Eucalyptus	Volumetric wood increase
Flax, Linseed	HT
Maize	Abiotic stress tolerance, HT, IR, pollination control system, lysine production, modified amylase production, phytase production
Melon	Delayed ripening
Papaya	Disease resistance
Plum	Disease resistance
Polish canola	HT
Poplar	IR
Potato	Disease resistance, HT, IR, modified starch/carbohydrate, reduced acrylamide potential, black spot bruise tolerance
Rice	HT, IR, anti-allergy
Soybean	HT, IR, modified oil/fatty acid, altered growth/yield
Squash	Disease resistance
Sugar beet	HT

Sugarcane	Drought stress tolerance
Sweet pepper	Disease resistance
Tobacco	HT, nicotine reduction
Tomato	Disease resistance, IR, delayed ripening, delayed fruit softening
Wheat	HT

Source: ISAAA GM Approval Database. http://www.isaaa.org/gmapprovaldatabase/default.asp

Biotech soybean

Herbicide tolerant (HT) soybean

HT soybean varieties contain a gene that provides resistance to one of two broad spectrum herbicides. This modified soybean has better weed control and reduced crop injury. It also improves farm efficiency by optimizing yield, using arable land more efficiently, saving time for the farmer, and increasing the flexibility of crop rotation. It encourages adoption of no-till farming, an important part of soil conservation practice. These varieties are the same as other soybeans in nutrition, composition, and in the way they are processed into food and feed.

Insect resistant (IR) soybean

This biotech soybean exhibits resistance to lepidopteran pests through the production of Cry1Ac protein. It was developed to reduce or replace high insecticide applications and at the same time maintain soybean yield potential.

Oleic acid soybean

This soybean contains high levels of oleic acid, a monounsaturated fat. According to health nutritionists, monounsaturated fats are considered "good" fats compared with saturated fats found in beef, pork, cheese, and other dairy products. Oil processed from these varieties is similar to that of peanut and olive oils. These new varieties have an oleic acid content that exceeds 80%, compared to only 24% from conventional soybeans.

Biotech maize

HT maize

These maize varieties work in a similar manner to HT soybean. They allow growers better flexibility in using certain herbicides to control weeds that damage crops.

IR maize

This modified maize contains a built-in insecticidal protein from *Bacillus thuringiensis* (Bt), a naturally occurring soil microorganism that gives maize plants protection from corn borers. Farmers do not have to spray insecticide to protect maize from pests. Bt maize also reduces toxin contamination from fungal attack on damaged grains. The Bt protein has been used safely as an organic insect control agent for over 50 years.

Biotech cotton

HT cotton
This cotton works in a manner similar to other HT crops.

IR cotton
This modified cotton contains a protein that provides the plant with season-long protection from budworms and bollworms. The need for additional insecticide applications for these pests is reduced or eliminated.

Biotech canola

HT canola
HT canola contains transgenes conferring tolerance to herbicides, similar to the trait exhibited by HT soybean.

High laureate canola
These canola varieties contain high levels of laurate. Oil processed from these varieties is similar to coconut and palm oils. This canola oil is being sold to the food industry for use in chocolate candy coatings, coffee whiteners, icings, frostings, and whipped toppings. Benefits extend even to the cosmetics industry.

Biotech potato

IR potato
This biotech potato contains a protein that provides the plant with built-in protection from the Colorado potato beetle. This variety needs no additional protection for this pest, benefiting farmers, consumers, and the environment.

VR potato
Several potato varieties have been modified to resist potato leafroll virus (PLRV) and potato virus Y (PVY). In the same way that people get inoculations to prevent disease, these potato varieties are protected through biotechnology from certain viruses. Furthermore, virus resistance often results in reduced insecticide use, which is needed to control insect vectors that transmit viruses.

Low acrylamide potato
Innate™ potato, developed by Simplot, was approved for commercialization in the U.S. in November 2014. This biotech potato has 50-75% lower levels of acrylamide, a potential carcinogen in humans, produced when potatoes are exposed to high temperatures. It is also less susceptible to bruising.

Biotech rice

HT rice
This rice contains a gene that provides resistance to one of two broad spectrum, environmentally benign herbicides.

Insect tolerant rice
This modified rice reduces yield losses caused by caterpillar pests, the most important of which are the yellow stem borer in tropical Asia and the striped stem borer in temperate areas.

Other biotech crops

Delayed ripening tomato
The delayed ripening (DR) tomato became the first GM food crop to be produced in a developed country. DR tomatoes spend more days on the vine, resulting in better flavor. These tomatoes have longer shelf life which is advantageous in harvesting and shipping due to reduced production costs.

HT sugar beet
In 2008, an HT sugar beet variety was planted in Canada and U.S. for the first time. HT sugar beet allows farmers to cut the number of required cultivations by half.

Virus resistant papaya
This Hawaiian-developed papaya contains a gene that encodes for the coat protein of papaya ringspot virus (PRSV). This protein provides the papaya plant with built-in protection against PRSV.

Virus resistant squash
A biotech yellow crookneck squash is able to resist watermelon mosaic virus (WMV) and zucchini yellow mosaic virus (ZYMV). This variety contains the coat protein genes of both viruses. This biotech approach bypasses aphid control, which reduces or eliminates the use of insecticides.

Non-browning apple
Two non-browning Arctic® apples, Arctic® Golden and Arctic® Granny, developed by Okanagan Specialty Fruits Inc., was approved for planting in the U.S. in February 2015. The non-browning Arctic® apples are identical to other apples, but do not turn brown when bruised, bitten, or cut.

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Global Status of Commercialized Biotech/GM Crops

In 2014, the global area of biotech crops continued to increase for the 19th year at a sustained growth rate of 3% to 4%, or 6.3 million hectares (Mhas) (~16 million acres), reaching 181.5 million hectares or 448 million acres (Figure 1). The area planted to biotech crops has grown impressively year after year for the past 19 years, with a remarkable 100-fold increase since commercialization began in 1996. Thus, biotech crops are considered as the fastest adopted crop technology in the history of modern agriculture.

In 2014, a total of 18 million farmers planted biotech crops in 28 countries, wherein over 94.1% (16.9 million) were small and resource-poor farmers from developing countries. The highest increase in any country, in absolute hectarage growth was in the U.S. with 3 million hectares.

During the period 1996 to 2014, biotech crops were successfully grown in an accumulated hectarage of 1.78 billion hectares (4.4 billion acres) around the world.

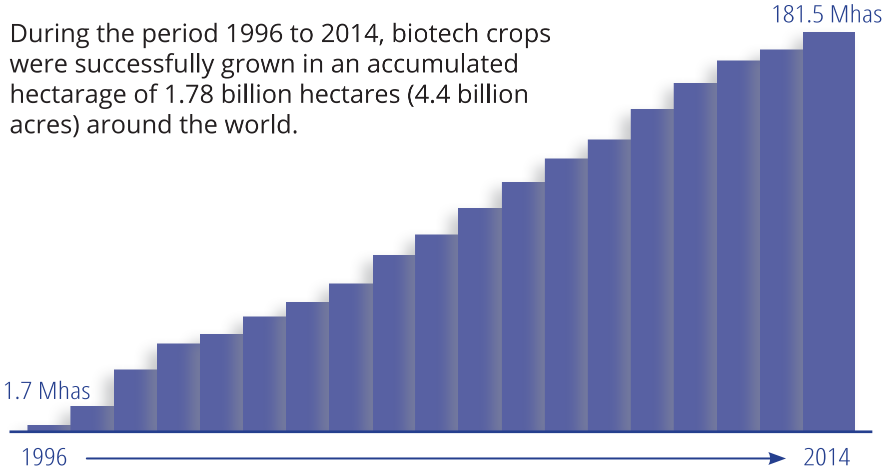

Figure 1. Global area of biotech crops, 1996 to 2014 (million hectares)

Distribution of biotech crops in industrial and developing countries

Figure 2 shows the area of biotech crops in industrial and developing countries from 1996-2014. In 2014, more than half (53%) of the global biotech crop area of 181.5 million hectares (96.2 million hectares), was in 20 developing countries. Unlike 2013, year-to-year growth was higher in the industrial countries at 4.2 million hectares (5%) than in developing countries at 2.1 million hectares equivalent to a 2% growth. This was principally due to higher growth in the U.S. (soybean) and Canada (canola) in 2014. Thus, whereas year-to-year growth was significantly faster in industrial countries in 2014, developing countries maintained a larger share of global biotech crops at 53% compared with only 47% for industrial countries.

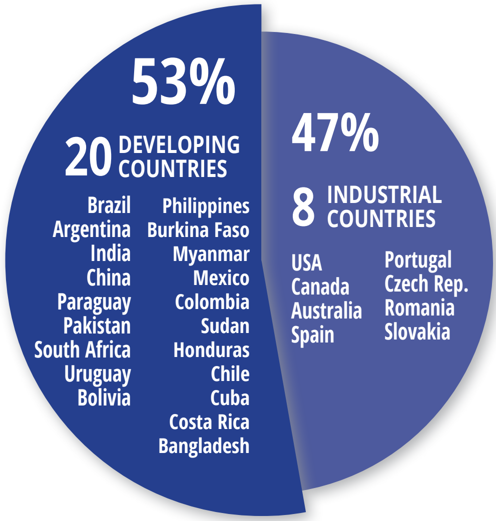

Figure 2. Distribution of biotech crops in industrial and developing countries

Global adoption of biotech soybean, maize, cotton, and canola

To provide a global perspective of the status of biotech crops, the global adoption rates as a percentage of the respective global areas of the four principal crops – soybean, cotton, maize and canola, is presented in Figure 3.

In 2014, 82% (90.7 million hectares) of the 111 million hectares of the soybean planted globally were biotech. Biotech cotton was planted to 25.1 million hectares, which is 68% of the 37 million hectares of global cotton. Of the 184 million hectares of global maize planted in 2014, 30% or 55.2 million hectares were biotech maize. Finally, herbicide tolerant biotech canola was planted in 9 million hectares or 25% of the 36 million hectares of canola grown globally in 2014. If the global areas (conventional and biotech) of these four crops are aggregated, the total area is 368 million hectares, of which 49% or 181.5 million hectares were biotech.

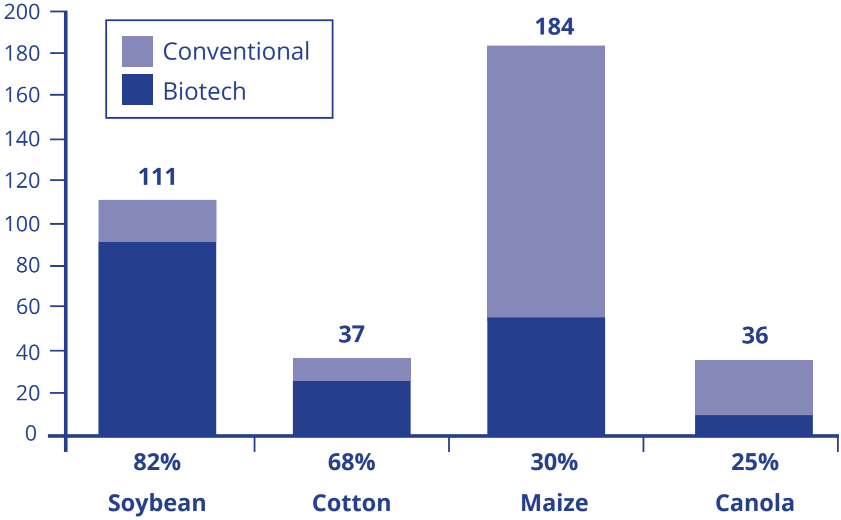

Figure 3. Biotech crop area as % of global area of principal crops, 2014 (million hectares)

The global value of biotech crops

In 2014, the global market value of biotech crops was US$15.7 billion representing 22% of the US$72.3 billion global crop protection market in 2013, and 35% of the ~US$45 billion global commercial seed market. Of the US$15.7 billion biotech crop market, US$11.3 billion (72%) was in the industrial countries and US$4.4 billion (28%) was in the developing countries. The market value of the global biotech crop market is based on the sale price of biotech seeds plus any technology fees that apply. The accumulated global value of biotech crops since 1996 is estimated at US$133,541 billion.

Future prospects

The biotech pipeline is filled with new crops and traits which could be commercialized in the next five years or more. These include products with multiple modes of resistance to pests and diseases, and tolerance to herbicides. Vitamin A-enriched rice (Philippines) and late blight resistant potatoes (Bangladesh, Indonesia, and India) field testings are progressing. The developer of Innate™ potato with late blight resistance and lowered reducing sugars has requested for approval in the U.S. In Africa, biofortified bananas and pest resistant cowpea look promising.

Biotech crops is not a panacea; but they have the potential to make a substantial contribution in cutting poverty by half, by optimizing crop productivity, which can be achieved by public-private sector partnerships.

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Note: Visit www.isaaa.org for annual global biotech status report.

Documented Benefits of GM Crops

The global area planted to GM crops has consistently increased over the past years. GM crops have been largely grown in developed countries. During the last few years, however, there has been a consistent increase in the number of developing countries planting GM crops.

Fifty-three percent (53%) of the total global GM crop area is now being grown in developing countries of Africa, Asia, and Latin America. Experiences from these countries show that resource-poor farmers can also benefit from this technology.

Global impact of GM crops

Farm Income

Biotech crops have had a positive impact on farm income worldwide due to enhanced productivity and efficiency gains. In 2013, direct global farm income benefit was $20.5 billion. Over the period of 18 years between 1996-2013, farm incomes have increased by $133.5 billion.[1]

Pesticide Use

Since 1996, farmers planting biotech crops have reduced pesticide inputs in their fields by 8.6%, or over 550 million kg which led to an overall reduction in the environmental footprint of biotech crops by 19%.

The volume of herbicides used by farmers planting herbicide tolerant (HT) maize has decreased by 210 million kg over the past 18 years. Similarly, significant reductions in pesticide loads were experienced by farmers planting insect resistant (IR) cotton and maize.[1]

Developed country experiences

United States

- An estimate cost savings by farmers planting HT soybean was $62/ha in 2013, almost three times higher compared to the early years of adoption. The annual total national farm income benefit from HT soybean has dramatically risen from $5 million in 1996 to $140 billion in 2013.[1]
- Glyphosate- and glufosinate-resistant corn reduced the herbicide use in corn production by 18.5 million pounds (15.2 and 3.3 million pounds, respectively) in 2004. US farmers saved $139 million from the reduced pesticide use.[2]
- The U.S. is estimated to have enhanced farm income from biotech crops by $58.4 billion in the period 1996 to 2013.[1]

Canada

- HT canola has boosted the total canola production in Canada by 11% in 2013. Adopters of biotech canola earned $546 million in 2013.[1]
- Canada is estimated to have enhanced farm income from biotech crops by $5.6 billion in the period 1996 to 2013.[1]

Australia

- For 2012, Australian farmers planting IR cotton have significant cost savings of about $186-270/ha despite the high cost of technology. In 2014, net farm income at the national level was $890 million.[3]

Developing country experiences

Bt cotton adoption in India

Cotton is a very important crop for India, accounting for 30% of its agricultural GDP. However, due to the high incidence of cotton bollworm, Indian farmers often lose up to 50-60% of their crop.[4] When Bt cotton was approved for commercial planting in India in 2002, bollworm infestation was suppressed.

In 2014, India topped biotech cotton production worldwide, planted on 11.6 million hectares, with 1,167 Bt cotton hybrids approved for planting.[3]

A study by Qaim and Khouser in 2013 investigated the effects of Bt cotton on 1,431 farm households in India. The results showed that adoption of Bt cotton has significantly improved calorie consumption and dietary quality, leading to increased family income. The technology reduced food insecurity by 15-20% among cotton-producing households.[5]

Biotech corn adoption in the Philippines

The approval of the commercial release of Bt corn in 2003 in the Philippines marked the first time that a GM food/feed crop was ever approved for planting in Asia. Plantings of Bt corn for the first year of commercial cultivation covered more than 10,000 hectares. Together with other biotech corn varieties (herbicide tolerant and Bt-HT), the total hectarage in the wet and dry seasons in 2014 was 831,000 hectares, up from 795,000 hectares in 2013.[3]

Adoption of Bt corn in the Philippines provided the following benefits to small-scale farmers:[3,6]

- Yield advantage of about 1.1 ton/ha or 30% yield increase over conventional corn hybrids
- Pesticide cost reduction by as much as 56%
- Profit gain of PhP10,132/ha (US$180), with PhP168/ha savings in insecticide costs
- Premium price for Bt corn because of good quality grains

Bt rice in China

Rice is the most important crop in China, with the highest level of production accounting for 28% of the world's total production.[7] In 2009, insect-resistant GM rice was approved for food, feed, and cultivation in China. To establish whether farmers' welfare improved by planting GM rice, surveys of farm households cultivating the biotech crop were conducted.

The surveys showed that small and poor farm households who adopted GM insect resistant rice had higher crop yields and lower pesticide usage compared to non-GM adopters. GM rice yields were 6% to 9% higher compared to conventional varieties and required less pesticide input by as much as 80% or 16.77 kg/ha, which contributed to improved health of farmers.[8]

Conclusion

The increasing number of farmers who are planting GM crops is a strong evidence of their advantages in agricultural production. In 2014, an accumulated hectarage of over 1.7 billion hectares of GM crops were planted by 18 million farmers. This unprecedented high adoption rate reflects the trust and confidence of millions of farmers in crop biotechnology.[3]

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Adoption and Uptake Pathways of Biotech Crops by Small-Scale Farmers in China, India, and the Philippines

GM crops have been offered as a modern crop development alternative to address the attack of pests and diseases, the vagaries of weather and other challenges to growing crops. Contrary to the notion that only farmers from developed countries are reaping the gains of biotechnology about 85% of these farmers are small landholders in the developing countries of China, India, and the Philippines.[1]

The *Adoption and Uptake Pathways of Biotech Crops by Small-Scale, Resource-Poor Asian Farmers: Comparative Studies in China, India, and the Philippines* project was spearheaded by ISAAA to give a human dimension to the statistics on farmer adoption and uptake pathways of GM crops and the changes these have brought about in resource-poor farmers' lives. "Adoption" refers to how farmers acquire and eventually apply the knowledge and practices pertaining to the planting of a GM crop and "uptake pathway" involves the process of capturing how a GM crop is introduced, adopted, spread, and shared by farmers with others.[2]

Collaborators from Chinese Academy of Sciences; the Indian Society for Cotton Improvement; and the University of the Philippines Los Baños surveyed farmers from China, India, and the Philippines.[2,3]

Who are the farmers using GM crops?

Traditionally, small-scale farming in developing countries has been stereotyped as backbreaking, unprofitable, and unappealing to the youth. But farmers planting biotech crops paint a different picture.

While Bt cotton production is still a male-dominated activity, there is growing involvement of women in GM crop commercialization in China. Based on focus group discussions, more women are attracted to the benefits of growing Bt cotton as there is less labor involved than would otherwise be needed for pesticide applications.

Filipino males dominate the planting process but wives control the finances and thus are major decision makers in the choice of crop to plant and farming methods to adopt. In Indian households, planting of

Bt cotton has become a family affair with the household head taking the more strenuous activities and mothers and children helping to pick and clean cotton bolls. In India, it is a significant sign that Bt cotton is attracting the young with over 50% in the 21-40 age bracket.

In the Philippines, even college graduates are venturing into GM maize production because they find it a viable income-generating alternative. Farmers in China and the Philippines report 2 to 3 times higher income from planting GM crops while Indian farmers obtain twice the income over traditional varieties.

Reasons for adoption of GM crops

Higher economic and yield benefits, freedom or reduced infestations from pests, and dramatic reduction in pesticide use are the principal motivators for adoption of GM crop in the three countries. Presence of private traders that sell seeds and provide capital loans as well as trust and strong ties among farmers that contributed to the information flow on GM crops also facilitated adoption.

As with any technology, there are also factors that limit or slow down adoption and uptake of GM crops. These include lack of capital and high cost of farm inputs especially in India and the Philippines. Influence of elders and church groups skeptical of GM crops in these two countries were also noted. In China, local seed companies could not meet the demand for GM seeds in the initial years of commercialization. Limited access to information about the new technology and inadequate government support also contributed to delayed adoption.

It is not government agricultural extension services that are crucial in farmer adoption. Rather, farmer leaders or village cadres have become local champions of GM crops as they take frontline action in trying out the technology after seeing a demonstration field trial, sharing their knowledge and signifying commitment to spread the benefits with fellow farmers.

Uptake pathways of GM crops

Field research indicates that early adopting farmers in India and the Philippines take the risk of trying out a GM crop which they initially heard about, from a demonstration field trial set up by seed companies, or from progressive village leaders. Other farmers take time to see how things progress, but become easily motivated to try the new crop after seeing convincing results of higher yields from the early adopters.

Early adopters are usually committed to sharing GM crop know-how with their relatives and peers. This is due to the prevailing strong peer system among farmers and the belief that they owe it to themselves and their fellow farmers to share what would benefit the community.

In China, village cadres coordinate with technicians to arrange training and convince farmers to participate in farm-related activities. Hence, the factors that facilitate early adoption are three-fold: support given by trusted village leaders for GM crop production; close ties among farmers; and avoidance of heavy losses incurred by farmers in cultivating non-GM crops.

Table 1. Facilitating factors in the adoption and uptake pathways of biotech crops

Nature	Facilitating Factors
Economic	• Financial benefits of cultivating GM crops (C, I, P) • Proof of good yield and income from early adoptors (C, I, P) • Presence of private traders of GM crops (C, I, P), providing loans for GM crop production (I, P), and buying harvests (C, I, P) • Availability of other financiers for GM crop production capital (P) • Financial losses from planting non-GM crops (C, P)
Political	• Cadres help to coordinate seminars and visits to demo fields (C) • "Breeding contract" between local seed companies and village chiefs for seed production (C) • Presence of farmer associations providing support (I, P)
Cultural	• Trust and strong ties among farmers (C, I, P) • Rapid spread of information on biotech crop (C, I, P)
Agriculture-related	• Synchronized farming (P) • Rapid spread of information on biotech crop (C)
Legend: C-China; I-India; P-Philippines	

Table 2. Limiting factors in the adoption and uptake pathways of biotech crops

Nature	Limiting Factors
Economic	• Lack of capital (I, P) • High cost of farm inputs (P) • Inadequate supply of GM seeds due to high demand (C) • Availability of seeds (P) • Low market price of harvests (P)
Political	• Indecisive local politicians (P)
Cultural	• Influence of elders skeptical of GM crops (I) • Influence of church groups who are against GMOs (P)
Agriculture-related	• Lack of land area for GM crop production (P) • Unsuitability of farm area for GM crop production (P) • Availability of alternative crops to plant (P) • Unfavorable weather conditions (I, P)
Communica-tional	• Lack of knowledge and misinformation about GM crops (C, I, P)
Legend: C-China; I-India; P-Philippines	

Conclusion

The champions of GM crops are the farmers. The farmers themselves decide whether to plant a crop or not, choose the variety to plant, and adopt new techniques and practices based on the benefits exhibited by progressive village leaders. During adoption, problems still arise which require the participation and cooperation of both the public and private sectors.

Farmer adoption of Bt cotton is now more than 95% of total cotton production in China and India while 80% of Filipino yellow corn farmers are planting GM maize.

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Are Foods Derived from GM Crops Safe?

GM crops are developed using modern biotechnology where precise tools are used to introduce only the desirable traits into a plant. In traditional plant breeding, however, genes from two parents are mixed in many different combinations to get the desired trait. Both methods have the potential to alter the nutritional value of plants, or lead to unintended changes in concentration of natural toxicants or anti nutrients. These concerns are less frequent in GM crops since only a limited number of genes are transferred during genetic modification.

Foods derived from GM crops have undergone more testing than any other food product in history. GM crops are assessed using guidelines issued by competent international scientific agencies such as the World Health Organization (WHO), the Food and Agriculture Organization (FAO), and the Organization for Economic Cooperation and Development (OECD). These guidelines include the following:

- GM food products should be regulated in the same way as foods produced by other methods.
- These products will be judged on their individual safety, allergenicity, toxicity, and nutrition rather than the methods or techniques used to produce them.
- Any new ingredient added to food through biotechnology will be subject to pre-market approval in the same way a new food additive, such as a preservative or food color, must be approved before it reaches the marketplace.

How are foods from GM crops assessed for food safety?

GM foods are exhaustively tested by the developer and independently evaluated for safety by experts in nutrition, toxicology, and allergenicity. The assessments are based on guidelines issued by competent regulatory agencies of each country.

Typical questions that must be addressed are:

- Does the GM food have a traditional counterpart that has a history of safe use?
- Has the concentration of any naturally occurring toxins or allergens in the food changed?
- Have the levels of key nutrients changed?
- Do new substances in the GM food have a history of safe use?
- Has the food's digestibility been affected?
- Has the food been produced using accepted, established procedures?

What are the issues?

Toxicity

Plants have low concentration of toxins to protect it from insect pests and diseases. The US Food and Drug Administration has guidelines that determine the normal and acceptable toxin levels of all crop varieties consumed based on toxicological studies. Natural toxin levels of GM crops are similar to their conventional counterparts.

Allergenicity

One of the public's biggest concerns related to GM foods is that an allergen could be accidentally introduced into a food product. There are 500 amino acid sequences of known protein allergens and 90% of all food allergies are associated with only eight foods or food groups — shellfish, eggs, fish, milk, peanuts, soybeans, tree nuts, and wheat. These, and many other food allergens are well characterized and so it is extremely unlikely that they would ever be introduced into a GM food.

The proteins introduced into commercially available GM foods are from sources with no history of allergenicity or toxicity; do not resemble known toxins or allergens biochemically and structurally; and their functions are well understood. DNA is present in all foods, and its ingestion is not associated with any ill effects. In fact, humans take in DNA every time they eat as it is present in all plant and animal material even whether it is cooked or raw.

Antibiotic resistance

Some GM crops contain antibiotic resistance genes to identify cells

into which the desired gene has been successfully introduced. Concerns have been raised that these marker genes could move from GM crops to microorganisms that normally reside in a person's gut and lead to an increase in antibiotic resistance. There have been numerous scientific reviews and experimental studies of this issue and they have come to the following conclusions:

- The likelihood of antibiotic resistance genes moving from GM crops to any other organisms is extremely remote or virtually zero.
- In the unlikely event that an antibiotic resistance gene is transferred to another organism, the impact would be negligible, as the markers used in GM crops have limited clinical or veterinary use.

Nevertheless, in response to public concerns, scientists have been advised to avoid using antibiotic resistance genes in GM plants.

Substantial equivalence (SE) in safety assessment of GM foods

Absolute safety is unattainable for any food product as people react differently to natural ingredients. Substantial equivalence (SE) is an alternative approach used for the safety assessment of genetically modified foods where traditional toxicological testing and risk assessment to whole foods could not be applied. It is based on the idea that existing products used as foods or food sources can serve as basis for comparison. The safety assessment is therefore based on a comparison of the modified food to its traditional (non-GM) counterpart in terms of molecular, compositional, toxicological and nutritional data. SE has been used in the safety assessment of GM crops available today.

Conclusion

Foods derived from GM plants are safe. Major issues and safety concerns on the biosafety of foods derived from GM plants have been addressed. Commercially available GM crops have passed through rigorous tests that showed they are non-toxic, non-allergenic, and their nutritional content is comparable to their non-GM counterparts.

The FAO, WHO, European Commission, French Academy of Medicine, American Medical Association, and American Society of Toxicology have come to an agreement that GM foods are safe for human health.

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GM Crops and the Environment

An increasingly growing population, global warming, and loss of biodiversity have a tremendous impact on the environment. By 2050, the world's population is estimated to be at 9.5 billion. Feeding these people will mean massive changes in the production, distribution, and stability of food products.

The destruction of wilderness and forests, and the continued use of coal and oil have led to a steady increase in carbon dioxide levels, resulting in global warming. It is predicted that the average global temperature will rise by 1.4 – 5.8ºC by 2100, with increasing fluctuations in weather conditions. Climate change will alter rainfall patterns and therefore require the migration of people and shifts in agricultural practices.

To conserve forests, habitats, and biodiversity, it is necessary to ensure that future food requirements come only from cropland currently in use.

What are the environmental benefits of GM crops?

One of the significant environmental benefits of GM crops is reduction in pesticide use, estimated at 37% in 2014, with the size of the reduction varying between crops and introduced trait.[1]

- A study assessing the global economic and environmental impacts of biotech crops for the first 18 years (1996-2013) of adoption showed that the technology has reduced pesticide spraying by 550 million kg and has reduced environmental footprint associated with pesticide use by 19%. The technology has also significantly reduced the release of greenhouse gas emissions from agriculture equivalent to removing 12.4 million cars from the road for a year.[2]
- In the U.S., adoption of GM crops resulted in pesticide use reduction of 110 million pounds in 2006.[3]
- The use of Bt cotton in China resulted in pesticide use reduction of 78,000 tons of formulated pesticides in 2001. This corresponds to about a quarter of all the pesticides sprayed in China in the

mid-1990s.[4] Additionally, the use of Bt cotton can substantially reduce the risk and incidence of pesticide poisonings to farmers.[5]

- The quantity of insecticides used to control bollworm reduced by 96% from 5,748 metric tons of active ingredients in 2001 to as low as 222 metric tons of active ingredients in 2011.
- Herbicide tolerant crops have facilitated the continued expansion of conservation tillage, especially no-till cultivation system, in the U.S. The adoption of conservation and no-till cultivation practices saved nearly 1 billion tons of soil per year.[6]
- Biotech cotton has been documented to have a positive effect on the number and diversity of beneficial insects in the U.S. and Australian cotton fields.[7]
- Adoption of Bt corn in the Philippines did not show an indication that Bt corn had negative effect on insect abundance and diversity.[8]

How are GM crops assessed for environmental safety?

GM crops undergo rigorous environmental safety assessment tests before they are approved for commerical cultivation. They are assessed by many stakeholders in accordance with principles developed by environmental experts around the world.[9,10,11] Among those who conduct risk assessment procedures are the developers of GM crops, regulatory bodies, and academic scientists.

Most countries use similar risk assessment procedures in considering the interactions between a GM crop and its environment. These include information about the role of the introduced gene, and the effect that it brings into the recipient plant. Also addressed are specific questions about unintentional effects such as:

- impact on non-target organisms in the environment
- whether the modified crop might persist in the environment longer than usual or invade new habitats
- likelihood and consequences of a gene being transferred unintentionally from the modified crop to other species

In addition to pre-commercialization tests for environmental safety, GM crops should also go through post approval monitoring by the product developer, independent researchers, and government scientists. This helps ensure that biotech crops continue to be safe for consumers and the environment.[12]

What are the potential risks?

Introduced genes outcrossing to weedy relatives as well as the potential to create weedy species

A major environmental concern associated with GM crops is their potential to create new weeds through outcrossing with wild relatives, or simply by persisting in the wild themselves. A ten-year study initiated in 1990 demonstrated that there is no increased risk of invasiveness or persistence in wild habitats for GM crops and traits tested when compared to their unmodified counterparts.[13]

Direct effects on non-target organisms

In May 1999, it was reported that pollen from *Bacillus thuringiensis* (Bt)-insect resistant corn had a negative impact on Monarch butterfly larvae. The report raised concerns about the potential risks to Monarch butterflies and other non-target organisms. Scientists, however, urged caution over the interpretation of the study because it reflects a different situation than that in the environment. A collaborative research by North American scientists concluded that in most commercial hybrids, Bt expression in pollen is low, and laboratory and field studies show no acute toxic effects at any pollen density that would be encountered in the field.[14] Laboratory experiments on predators and extensive field work demonstrated no significant impact on Monarch Butterfly populations.[12]

Development of insect resistance

Another concern over the use of Bt crops is the development of insect resistance to Bt. Insect resistance management plans have been developed by government, industry, and scientists to address this issue.

Conclusion

The environmental concerns potentially associated with GM crops are evaluated prior to their release for commercial planting. Post-approval monitoring and good agricultural systems ensure that GM crops continue to be safe after their release.

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Biotechnology and Climate Change

Increasing greenhouse gas emissions has raised the temperature of the earth's atmosphere. This results to melting of glaciers, unpredictable rainfall patterns, and extreme weather events. The accelerating pace of climate change, combined with global population and depletion of agricultural resources, threatens food security globally.

In 2012, almost 40% of the world population of 6.7 billion, equivalent to 2.5 billion, rely on agriculture for their livelihood and will thus likely be the most severely affected.[1] To mitigate these effects, current agricultural approaches need to be modified and innovative adaption strategies need to be in place to efficiently produce more food in stressed conditions and with net reduction in greenhouse gas emissions.

Contribution of biotech crops in mitigating effects of climate change

Green biotechnology can help decrease greenhouse gases and therefore mitigate climate change. Since their commercialization in 1996, biotech crops have been contributing to the reduction of CO2 emissions. They allow farmers to use less and environmentally friendly energy and fertilizer, and practice soil carbon sequestration.

- Herbicide tolerant biotech crops such as soybean and canola facilitate zero or no-till, significantly reducing the loss of soil carbon, CO2 emissions, fuel use, and soil erosion.
- Insect resistant biotech crops require fewer pesticide sprays resulting in savings of tractor/fossil fuel and thus less CO2 emissions. In 2013, the combined permanent and additional savings through carbon sequestration was equivalent to a saving of 28 billion kg of CO2, or removing 12.4 million cars off the road.[2]

Biotech crops adapted to climate change

Crops can be modified faster through biotechnology. Pest and disease resistant biotech crops have been continuously developed as new pests and diseases emerge while climate changes. Insect resistant varieties reduce pesticide applications and crops tolerant to various abiotic stresses have been developed in response to climatic changes.

Salinity tolerant crops

Salt tolerant crops have been developed and some are in the final field trials before commercialization. In Australia, field trials of 1,161 lines of genetically modified (GM) wheat and 1,179 lines of GM barley modified to contain one of 35 genes obtained from wheat, barley, maize, thale cress, moss, or yeasts are in progress since 2010. Some of the genes are expected to enhance tolerance to a range of abiotic stresses including drought, cold, salt, and low phosphorous. Sugarcane that contains transcription factor (OsDREB1A) is also under field trial since 2009.[3]

More than a dozen of other genes for salt tolerance have been found in various plants. Some of these candidate genes may prove feasible in developing salt tolerance in sugarcane[3], rice[4,5], barley[4], wheat[6], tomato[7], and soybean.[8]

Drought tolerant crops

Transgenic plants carrying genes for water-stress management have been developed. Structural genes (key enzymes for osmolyte biosynthesis, such as proline, glycine/betaine, mannitol and trehalose, redox proteins and detoxifying enzymes, stress-induced LEA proteins) and regulatory genes, including dehydration–responsive, element-binding (DREB) factors, zinc finger proteins, and NAC transcription factor genes, are being used. Transgenic crops carrying different drought tolerant genes are being developed in rice, wheat, maize, sugarcane, tobacco, Arabidopsis, groundnut, tomato, potato and papaya.[9,10]

An important initiative for Africa is the Water Efficient Maize for Africa (WEMA) project of the Kenyan-based African Agricultural Technology Foundation (AATF) and funded by the Bill and Melinda Gates Foundation (BMGF) and Howard G. Buffet Foundation. Drought tolerant WEMA varieties developed through marker assisted breeding could be available to farmers within the next two or three years. Drought tolerant and insect-protected varieties developed using both advanced breeding and transgenic approaches could be available to farmers in the later part of the decade.[11] In 2012, a GM drought tolerant maize MON 87460 that expresses cold shock protein B has been approved in the U.S. for release in the market.[12]

Biotech crops for cold tolerance

Through genetic and molecular approaches, a number of relevant genes have been identified. These include the genes controlling the *CBF* cold-responsive pathway and together with *DREB1* genes, integrate several components of the cold acclimation response to tolerance low temperatures.[13]

Cold tolerant GM crops are being developed such as GM eucalyptus, which is currently being field tested in the U.S. by Arborgen LLC. Thale cress has been improved to contain the *DaIRIP4* from *Deschapsia antarctica*, a hairgrass that thrives in frosts down to -30^{O}C, and sugarcane are being introgressed with genes from cold tolerant wild varieties.[3]

Biotech crops for heat stress

Heat shock proteins (HSPs) has been associated with recovery of plants under heat stress and sometimes, even during drought. HSPs bind and stabilize proteins that have become denatured during stress conditions, and provide protection to prevent protein aggregation. In GM chrysanthemum with the *DREBIA* gene from *Arabidopsis thaliana*, the transgene and other heat responsive genes such as the HSP70 (heat shock proteins) were highly expressed when exposed to heat treatment. The transgenic plants maintained higher photosynthetic capacity and elevated levels of photosynthesis-related enzymes.[14]

Forward looking

Improved crops resilient to extreme environments are expected in a few years. Biotech research to mitigate global warming should be initiated to sustain the utilization of new products. The induction of nodular structures on the roots of non-leguminous cereal crops to fix nitrogen will reduce farmers' reliance on inorganic fertilizers. The utilization of excess CO2 in the air by staple crop rice by converting its CO2 harnessing capability from C3 to C4 pathway. C4 plants like maize can efficiently assimilate and convert CO2 to carbon products during photosynthesis.

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GM Crop Traits

Bt Insect Resistance Technology

The Bt organism

Bt stands for *Bacillus thuringiensis* (Bt) a common soil bacterium that was first isolated in Thuringia, Germany. Bt produces a protein that paralyzes the larvae of some harmful insects, including the cotton bollworm and the Asian and European corn borers, which are common plant pests causing devastating effects on important crops.

Mode of action

When ingested by the larva of the target insect, the Bt protein is activated in the gut's alkaline condition and punctures the mid-gut leaving the insect unable to eat. The insect dies within a few days (Figure 1).

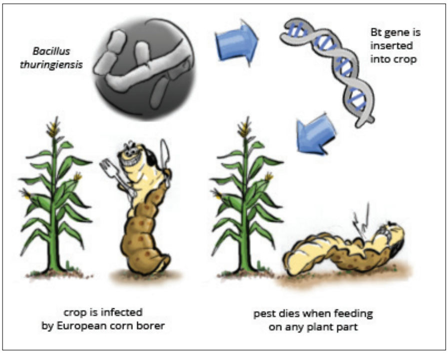


Figure 1. Bt's mode of action

It is because of its ability to produce the insecticidal protein that much research is being done to exploit the organism's agronomic value. There are more than 200 types of Bt proteins identified with varying degrees of toxicity to some insects.

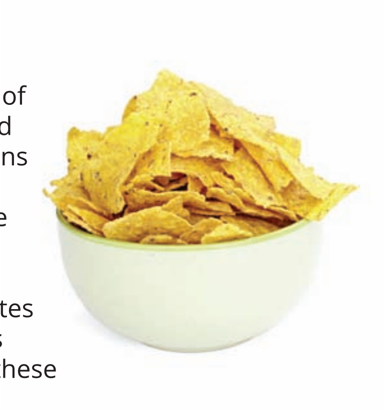
Earlier Bt technology

Bt is easily cultured by fermentation. For over 50 years, Bt has been used as an insecticide by farmers worldwide, even those who practice organic farming. The insecticide is applied either as a spray, or as ground application. However, the efficiency of both applications is quite limited, as target organisms often do not come in contact with the insecticide as they are found on the underside of leaves or have already penetrated into the plant. Scientists are working to overcome this problem through the use of modern biotechnology.

Modern Bt technology

Scientists have taken the Bt gene responsible for the production of the insecticidal protein from the bacterium and incorporated it into the genome of plants. Thus, these plants have a built-in mechanism of protection against targeted pests round the clock. The protein produced by the plants does not get washed away.

Safety aspects of Bt technology



Effects on human health

The specificity of Bt for its target insects is one of the characteristics that make it an ideal method of biological pest control. In fact, different strains of Bt have specific toxicity to certain target insects. The specificity rests on the fact that the toxicity of the Bt protein is receptor-mediated. This means that for an insect to be affected by the Bt protein, it must have specific receptor sites in its gut where the proteins can bind. Humans and majority of beneficial insects do not have these receptors.

Before Bt crops are placed on the market, they must pass very stringent regulatory tests, including those for toxicity and allergenicity.

The U.S. Environmental Protection Agency (USEPA) has already administered toxicology assessments, and Bt proteins have already been tested even at relatively higher dosages. According to the Extension Toxicology Network (Extoxnet), a pesticide information project of several American universities, “no complaints were made after 18 humans ate one gram of commercial Bt preparation daily for five days, on alternate days... Humans who ate one gram per day for three consecutive days were not poisoned or infected.” Furthermore, the protein was shown to be degraded rapidly by human gastric fluid *in vitro*.[1]

Effects on the environment

Soil ecosystems and groundwater

The Bt protein is moderately persistent in soil and is classified as immobile, as it does not move, or leach, with groundwater. It does not particularly persist in acidic soil conditions and, when exposed to sunlight, is rapidly destroyed due to UV radiation.

Independent experts have conducted studies to investigate the impact of Bt crops on soil organisms and other beneficial insects. No adverse effects have been found on non-target soil organisms, even when these organisms were exposed to quantities of Bt far higher than actual concentration in natural crop-growing conditions. Likewise, research done by the USEPA revealed no changes in the soil microbiota in fields with Bt plant material or conventional plant material[2] or between fields of Bt and non-Bt crops.[3]

Animals and insects

Monarch caterpillar

On tests conducted on dogs, guinea pigs, rats, fish, frogs, salamanders, and birds, the Bt protein was found not to have any harmful effects. No toxic effects were found on beneficial or predator insects, such as honeybees and lady beetles.[1]

In 1999, it was reported that pollen from Bt corn had a negative impact on Monarch butterfly larvae. This report raised concerns and questions about the risks of Bt crops on non-target organisms. Recent studies, however, show that Bt corn poses "negligible" threat to Monarch butterflies in the field. A collaborative research effort by scientists in the U.S. and in Canada has produced information to develop a formal risk assessment of the impact of Bt corn on Monarch butterfly populations. They concluded that in most commercial hybrids, Bt expression in pollen is low, and laboratory and field studies show no acute toxic effects at any pollen density that would be encountered in the field.[4]

Advantages of Bt crops

- Improved pest management
- Reduction in insecticide use
- Greater net return
- Improved conditions for non-target organisms
- Less mycotoxin in corn

Insect resistance management

Since Bt crops are capable of season long expression of the Bt protein, precautionary steps have to be taken in order to avoid the development of insect resistance. In the U.S., for example, USEPA usually requires a "buffer zone," or a structured refuge of non-Bt crops that is planted in close proximity to the Bt crops.[5] Insect resistance management (IRM) is said to be the key to sustainable use of the insecticide in both GM crops and Bt microbial spray formulations.

Conclusion

Bt crops are an addition to our arsenal against plant pests. With an increasing population and decreasing arable land, it is necessary to exploit all options with as little compromise to produce more crops. When used side by side with proper agricultural practices, Bt insect resistance technology can bring many benefits to crops, farmers, and consumers alike.

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Herbicide Tolerance Technology: Glyphosate and Glufosinate

Weeds are a constant problem in farmer's fields. Weeds not only compete with crops for water, nutrients, sunlight, and space but also harbor insects and diseases; clog irrigation and drainage systems; undermine crop quality; and deposit weed seeds into crop harvests. If left uncontrolled, weeds can reduce crop yields significantly.

Farmers can fight weeds with tillage, hand weeding, herbicides, or typically a combination of all techniques. Unfortunately, tillage leaves valuable topsoil exposed to wind and water erosion, a serious long-term consequence for the environment. For this reason, more farmers prefer reduced or no-till methods of farming.

Similarly, many have argued that the heavy use of herbicides has led to groundwater contaminations, the death of several wildlife species and has also been attributed to various human and animal illnesses.

Weed control practices

The tandem technique of soil-tilling and herbicide application is an example of how farmers control weeds in their farms.

Generally, they till their soil before planting to reduce the number of weeds present in the field. Then they apply broad-spectrum or non-selective herbicides (one that can kill all plants) to further reduce weed growth just before their crop germinates. This is to prevent their crops from being killed together with the weeds. Weeds that emerge during the growing season are controlled using narrow-spectrum or selective herbicides. Unfortunately, weeds of different types emerge in the field, and therefore, farmers have to use several types of narrow-spectrum herbicides to control them. This weed control method can be very costly and can harm the environment.

Researchers postulated that weed management could be simplified by spraying a single broad-spectrum herbicide over the field anytime during the growing season.

Development of glyphosate and glufosinate herbicide tolerant plants

Herbicide tolerant (HT) crops offer farmers a vital tool in fighting weeds and are compatible with no-till methods, which help preserve topsoil. They give farmers the flexibility to apply herbicides only when needed, to control total input of herbicides and to use herbicides with preferred environmental characteristics.

Technology background

How do these herbicides work?
These herbicides target key enzymes in the plant metabolic pathway, which disrupt plant food production and eventually kill it. So how do plants elicit tolerance to herbicides? Some may have acquired the trait through selection or mutation; or more recently, plants may be modified through genetic engineering.

Why develop HT crops?
Glyphosate and glufosinate herbicides are useful for weed control and have minimal direct impact on animal life, and are not persistent. They are highly effective and among the safest of agrochemicals. Unfortunately, they are equally effective against crop plants. Thus, HT crops are developed to have a degree of tolerance to these herbicides.

How do glyphosate and glufosinate HT crops work?

Glyphosate tolerant crops
Glyphosate kills plants by blocking the EPSPS enzyme, an enzyme involved in the production of aromatic amino acids, vitamins and many secondary plant metabolites. There are several ways by which crops can be modified to be glyphosate tolerant. One strategy is to incorporate a soil bacterium gene that produces a glyphosate tolerant form of EPSPS. Another way is to incorporate a different soil bacterium gene that produces a glyphosate degrading enzyme.

Glufosinate tolerant crops
Glufosinate herbicides contain the active ingredient phosphinothricin, which kills plants by blocking the enzyme responsible for nitrogen metabolism and for detoxifying ammonia, a by-product of plant metabolism. Crops modified to tolerate glufosinate contain a bacterial

gene that produces an enzyme that detoxifies phosphonothricin and prevents it from doing damage.

Other methods by which crops are genetically modified to survive exposure to herbicides including: 1) producing a new protein that detoxifies the herbicide; 2) modifying the herbicide's target protein so that it will not be affected by the herbicide; or 3) producing physical or physiological barriers preventing the entry of the herbicide into the plant. The first two approaches are the most common ways scientists develop herbicide tolerant crops.

Safety aspects of herbicide tolerance technology

Toxicity and allergenicity

Government regulatory agencies in several countries have ruled that HT crops do not pose any other environmental and health risks as compared to their non-GM counterparts.

Introduced proteins are assessed for potential toxic and allergenic activity in accordance with guidelines developed by concerned international organizations. They are from sources with no history of allergenicity or toxicity; they do not resemble known toxins or allergens; and they have functions, which are well understood.

Effects on the plants

The expression of these proteins does not damage the plant's growth nor result in poorer agronomic performance compared to parental crops.

Persistence or invasiveness of crops

A major environmental concern associated with herbicide tolerant crops is their potential to create new weeds through outcrossing with wild relatives or simply by persisting in the wild themselves. This potential, however, is assessed prior to introduction and is also monitored after the crop is planted. The current scientific evidence indicates that, in the absence of herbicide applications, HT crops are no more likely to be invasive in agricultural fields or in natural habitats than their non-GM counterparts.

The HT crops currently in the market show little evidence of enhanced persistence or invasiveness.

Advantages of herbicide tolerant crops

- Excellent weed control and hence higher crop yields;
- Flexibility – possible to control weeds later in the plant's growth;
- Reduced numbers of sprays in a season;
- Reduced fuel use (because of less spraying);
- Reduced soil compaction (because of less need to go on the land to spray);
- Use of low toxicity compounds which do not remain active in the soil; and
- Use of no-till or conservation-till systems

A study conducted by the American Soybean Association (ASA) on tillage frequency on soybean farms showed that significant numbers of farmers adopted the "no-tillage" or "reduced tillage" practice after planting HT soybean varieties. This simple weed management approach saved over 234 million gallons of fuel and left 247 million tons of irreplaceable topsoil undisturbed.

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Delayed Ripening Technology

Ripening is a normal phase in the maturation process of fruits and vegetables. Upon its onset, it only takes about a few days before the fruit or vegetable is considered inedible. This unavoidable process brings significant losses to both farmers and consumers alike.

Scientists have been working to delay fruit ripening so that farmers will have the flexibility in marketing their goods and ensure consumers of "fresh-from-the-garden" produce.

The fruit ripening process

Ethylene is a natural plant hormone associated with the growth, development, ripening and aging of many plants. This phytohormone is said to promote ripening in a variety of fruits including bananas, pineapples, tomatoes, mangoes, melons, and papayas. It is produced in varying quantities depending on the type of fruit. But when the concentration of ethylene reaches 0.1-1.0 ppm (parts per million), the ripening process in climacteric fruits is considered irreversible.

Climacteric fruits are usually harvested once they have reached maturity which then undergoes rapid ripening during transit and storage. Important tropical fruits such as banana, mango, papaya, pineapple and guava are examples of these fruits. Non-climacteric fruits do not ripen after harvest. Thus, in order to attain full ripeness and flavor, these fruits such as strawberries and oranges, are often harvested once they have fully ripened.

In tomatoes, it takes about 45-55 days for the fruit to reach full maturity. After which, it starts to undergo the ripening process. The production of ethylene within the fruit in turn signals the activity of different enzymes resulting in physiological changes such as the change of color from green to red, the softening of the fruit, and the development of its distinct taste and aroma.

Normally, farmers pick their produce while they are still green. The ripening process is then induced by spraying the fruits or vegetables with

ethylene gas when they reach their destination. For long hauls, fruits and vegetables are refrigerated to prevent damage and delay their ripening.

However, there are drawbacks to these postharvest practices. Fruits that have been harvested prematurely may result in poor taste and quality despite appearing as fully ripened ones. Fruits transported for long periods under refrigeration also have the tendency to lose their quality.

Controlling the ripening process

There are several ways by which scientists can control the ripening process by genetic modification.

Regulation of ethylene production

The amount of ethylene produced can be controlled primarily by "switching off" or decreasing the production of ethylene in the fruit and there are several ways to do this. They include:

- ***a. Suppression of ACC synthase gene expression.*** ACC (1-aminocyclopropane-1-carboxylic acid) synthase is the enzyme responsible for the conversion of S-adenosylmethionine (SAM) to ACC; the second to the last step in ethylene biosynthesis. Enzyme expression is hindered when an antisense ("mirror-image") or truncated copy of the synthase gene is inserted into the plant's genome.
- ***b. Insertion of the ACC deaminase gene.*** The gene coding for the enzyme is obtained from *Pseudomonas chlororaphis*, a common nonpathogenic soil bacterium. It converts ACC to a different compound thereby reducing the amount of ACC available for ethylene production.
- ***c. Insertion of the SAM hydrolase gene.*** This approach is similar to ACC deaminase wherein ethylene production is hindered when the amount of its precursor metabolite is reduced; in this case SAM is converted to homoserine. The gene coding for the enzyme is obtained from *E. coli* T3 bacteriophage.
- ***d. Suppression of ACC oxidase gene expression.*** ACC oxidase is the enzyme which catalyzes the oxidation of ACC to ethylene, the last step in the ethylene biosynthetic pathway. Through anti-sense technology, down regulation of the ACC oxidase gene results in the suppression of ethylene production, thereby delaying fruit ripening.

Control of Ethylene Perception

Since ethylene signals the onset of fruit ripening, delayed ripening on some plants can be achieved by modifying their ethylene receptors. The gene ETR1 is one example, and it has been shown to encode an ethylene binding protein. Plants with modified ETR1 lack the ability to respond to ethylene.

Suppression of Polygalacturonase Activity

Polygalacturonase (PG) is the enzyme responsible for the breakdown of pectin, the substance that maintains the integrity of plant cell walls. Pectin breakdown occurs at the start of the ripening process resulting in the softening of the fruit. To produce a fruit with DR trait using this method, scientists insert an anti-sense or a truncated copy of the PG gene into the plant's genome resulting in a dramatic reduction of the amount of PG enzyme produced thereby delaying pectin degradation.

Advantages of DR technology

The increased shelf life of products offers several advantages to both producers and consumers:

- *Assurance of top quality fruits and vegetables on the market.* Farmers can now wait for the fruits and vegetables to attain full maturity before they are plucked from their vines thereby allowing the fruits to exude full quality. Consumers will get value for their money.
- *Widening of market opportunities for farmers.* Produce can now be transported for longer periods of time, some of which would not even require refrigeration.

- *Reduction in postharvest losses.* DR fruits do not go soft easily compared to conventional ones and are therefore more resilient to damage during handling and transportation. This ensures a significant percentage of the harvested fruits to end up on the market shelves.
- *Extension in shelf life.* Fruits or vegetables as they stay fresher and nutritious for longer periods. These fruits will not easily go "over the hill".

Safety aspects of DR technology

The first ever GM crop approved for marketing was the Flavr-Savr™ tomato produced by Calgene, Inc. (U.S.) in 1994. After thoroughly studying DR technology and its products, U.S. regulatory agencies concluded that the DR technology is safe, it produces tomatoes that have the same nutritional composition as the conventional ones and that show no difference in levels of allergens or toxins compared to normal fruit. In addition, field trials have shown that the DR tomatoes do not pose any threat to other plants nor to any non-target organisms.

Other DR tomatoes that followed thereafter have also been granted deregulated status by regulatory agencies in several countries including the U.S., Canada, and Mexico. In 1996, the UK's food safety regulators also gave their thumbs up to a DR tomato developed by Zeneca Seeds but it is not currently being sold in supermarkets.

DR technology has been applied for use in tomatoes, melons, and papaya. DR technology is also used in carnation to delay the withering of flowers.

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Stacked Traits in Biotech Crops

Gene stacking (also known as gene pyramiding and multigene transfer) refers to the process of combining two or more genes of interest into a single plant. The combined traits resulting from this process are called stacked traits. A biotech crop variety that bears stacked traits is called a biotech stack or simply stack.

Compared to mono-trait crop varieties, stacks offer broader agronomic enhancements that allow farmers to meet their needs under complex farming conditions. Biotech stacks are engineered to have better chances of overcoming the myriad of problems in the field such as insect pests, diseases, weeds, and environmental stresses so that farmers can increase their productivity.

Gene stacking enhances and simplifies pest management for biotech crops as demonstrated by multiple insect resistance based on Bt technology. Experience has shown that the resistance conferred by a single Bt gene has the potential to break down as the target insect pest mutates and adapts to defeat the Bt trait.

To prevent or delay the emergence of resistance to the Bt gene, many regulatory agencies require a refuge or an area planted to a non-Bt variety alongside the Bt crop. Typically, a refuge is about 20 percent of the total crop area for a mono-Bt trait variety (Figure 1).

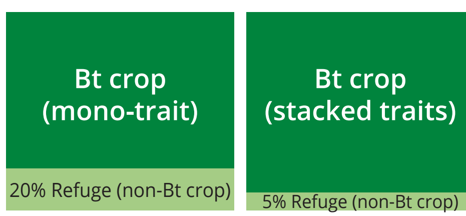

Figure 1. Refuge requirements for Bt crops

While the refuge strategy lessens the chance for the insect pest to overcome the Bt trait, farmers cannot realize the full production benefit of the Bt crop. The next generation of Bt crops with multiple modes of action for insect control were then developed by stacking several classes of Bt genes. This gene stacking approach has reduced the potential of resistance breakdown as it is more difficult for the pest to overcome multiple insecticidal proteins. This greater durability of Bt stacks allow a lower refuge area requirement that somehow limits yield.[1]

To catch up in countering weed resistance, biotech seed developers have stacked up genes to broaden the herbicidal mode of actions. For example, this is done by combining the glyphosate resistance gene *epsps* with the *pat* gene conferring resistance to herbicide glufosinate and/or with the *dmo* gene conferring resistance to herbicide dicamba.

Gene stacking is especially useful in metabolic engineering of plants since most metabolic processes and biochemical pathways involve numerous genes interacting with each other.[2] For example, the entire pathway for provitamin A (beta carotene) biosynthesis was engineered in the rice endosperm by stacking three carotenoid genes into rice.[3] The biotech rose with modified flower color was produced by stacking two genes in the anthocyanin biosynthetic pathway that altered the flower pigmentation process, giving the biotech rose flowers novel shades of blue.[4]

Gene stacking process

Table 1 summarizes the common methods of gene stacking and provides examples of commercialized stacks produced by each method.

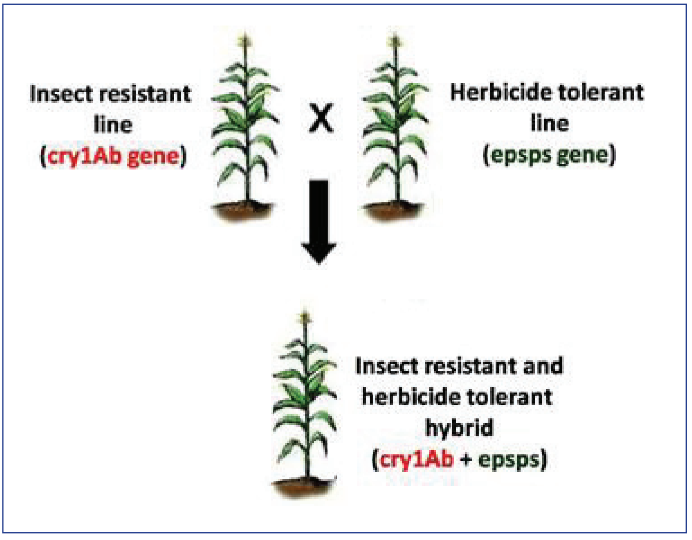

Figure 2. Stacking insect resistance and herbicide tolerance genes

Table 1. Comparison of most commonly used marker systems (adopted from Korzun, 2003)

Method	Description	Examples of commercial stacks*
Hybrid stacking	A plant harboring one or more transgenes is cross-hybridized with another plant containing other transgenes.	Cotton: Roundup Ready™ Flex Bollgard™ II
Co-transformation	A plant is transformed with two or more independent transgenes. The transgenes of interest are in separate gene constructs and delivered to the plant simultaneously.	Maize: YieldGard™
Linked genes/ multigene cassette transformation	A plant is transformed with a single gene construct that harbors two or more linked transgenes.	Soybean: Vistive™ Gold
Re-transformation	A plant harboring a transgene is transformed with other transgenes.	Cotton: Bollgard™ II

*Source: ISAAA GM Approval Database[5]

Regulatory approaches

Regulatory principles and procedures for approval and release of biotech stacks differ globally. In countries like the U.S. and Canada, no separate or additional regulatory approval is necessary for commercializing hybrid stacks that are products of crossing a number of already approved biotech lines. This policy is based on the argument that interactions between individual trait components in a stack that have been shown to pose no environmental or health hazard would not result in new or altered hazards. The U.S. Environmental Protection Agency, however, may require separate safety review of a stack upon identification of a specific hazard associated with combined "plant incorporated protectants" (PIPs), since combinations of PIPs may result in altered toxicity.

In Japan and European Union countries, stacks are considered new events, even if individual events have market approval, and must pass through a separate regulatory approval process, including risk assessment of their safety, similar to mono-trait biotech events. Risk assessment is focused on the identification of additional risks that could arise from the combined genes. Possible risks are altered effects of interacting proteins on the target and non-target organisms and increased invasiveness of the crop that may pose environmental risks.

Technological challenges

For the developer, the choice between a large molecular stack and a complex hybrid stack will be based on the monetary cost and timeline for developing and registering a stack. In countries where a stack must pass a separate regulatory review, the one-shot molecular stacking may be more cost effective than the lengthy hybrid stacking. There are, however, technological concerns in molecular stacking which include the design of large multi-gene constructs, method of delivery into plant cells and the stability of expression of multiple genes. Molecular biologists are developing new genetic engineering approaches to address these concerns. Among the promising technologies include site-specific gene recombination systems in conjunction with the use of engineered DNA cutting enzymes and the artificial gene assembly known as minichromosome.[6,7]

How the multiple transgenes might affect the overall physiology of the plant and how many genes and what combinations of genes can be stacked into a plant are of primary concern to developers. If multiple transgene insertions will alter protein synthesis and metabolic processes of the plant drastically, these likely will compromise the yield. While this yield drag is not necessarily a biosafety concern, the yield loss will need to be offset by reduced production cost and the extra premium farmers will make from the added value of the biotech trait(s) in order for a stack to be a viable and beneficial biotech product.

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Biotechnology for the Development of Drought Tolerant Crops

Water and agriculture

Adverse environmental factors, of which water scarcity represents the most severe constraint to agriculture, account for about 70 percent of potential yield loses worldwide.[1] It is thus essential to improve water use efficiency in agriculture. This will require an integrated approach to water resources management to encourage an efficient and equitable use of the resource, and to ensure sustainability. The development of crop varieties with increased tolerance to drought, both by conventional breeding methods and by genetic engineering, is also an important strategy to meet global food demands with less water.

Developing drought tolerant crops

Conventional breeding requires the identification of genetic variability to drought among crop varieties, or among sexually compatible species, and introducing this tolerance into lines with suitable agronomic characteristics. Conventional breeding for drought tolerance is a slow process because it is limited by the availability of suitable genes for breeding. The development of tolerant crops by genetic engineering, on the other hand, requires the identification of key genetic determinants underlying stress tolerance in plants, and introducing these genes into crops. Drought triggers a wide array of physiological responses in plants, and affects the activity of a large number of genes: gene expression experiments have identified several hundred genes which are either induced or repressed during drought.[2]

Plant drought tolerance mechanisms

Genetic control of tolerance to abiotic stresses is not only very complex, but is also highly influenced by other environmental factors and by the developmental stage of the plant.

The physiological responses of plants to a deficit of water include leaf wilting, a reduction in leaf area, leaf abscission, and the stimulation of root growth by directing nutrients to the underground parts of the plants. Plants are more susceptible to drought during flowering and seed development (the reproductive stages), as plant's resources are deviated to support root growth. In addition, abscisic acid (ABA), a plant stress hormone, induces the closure of leaf stomata (microscopic pores involved in gas exchange), thereby reducing water loss through transpiration, and decreasing the rate of photosynthesis. These responses improve the water-use efficiency of the plant on the short term.

Plant cells are required to maintain water balance. To mantain this water balance, plants absorb water when water potential is negative. Cells can decrease their water potential through the accumulation of solutes, such as sugars, amino acids, organic acids and ions – especially potassium (K+). As cellular enzymes are severely inhibited by the presence of ions, these must be removed from the cytosol (the ground fluid substance of the cell) and stored in special storage cell organelles, the vacuoles. Compatible solutes that accumulate in the cytosol and do not interfere with enzymatic reactions comprise sugar alcohols (mannitol and sorbitol), the amino acid proline, and glycine betaine. The synthesis of these compounds by the plant enhances tolerance to drought.[3]

The plant's response to drought is accompanied by the activation of genes involved in the perception of drought stress and in the transmission of the stress signal. One group are genes that encode proteins that protect the cells from the effects of desiccation. These genes include those that govern the accumulation of compatible solutes; passive transport across membranes; energy-requiring water transport systems; and protection and stabilization of cell structures from desiccation and damage by reactive oxygen species.[3]

A second group of genes activated by drought is comprised by regulatory proteins that further regulate the transduction of the stress signal and modulate gene expression. At least four independent stress-responsive genetic regulatory pathways are known to exist in plants, forming a highly complex and redundant gene network.[3,4] Two of the pathways are dependent on the hormone ABA, and two are ABA-independent. These pathways are also implicated in the perception and response to additional stress factors, including cold, high temperature and salinity.

Genetic engineering drought tolerant plants

Many of the genes known to be involved in stress tolerance have been isolated initially in Arabidopsis. The introduction of several stress-inducible genes into plants by genetic engineering has resulted to increased tolerance of transgenics to drought, cold and salinity stresses.[3,4]

Genetic manipulation of the stress response to abscisic acid (ABA)

ABA levels in the plant greatly increase in response to water stress, resulting in the closure of stomata thereby reducing the level of water loss through transpiration from leaves and activate stress response genes. The reaction is reversible: once water becomes available again, the level of ABA drops, and stomata re-opens. Increasing the plant's sensitivity to ABA has therefore been a very important target for improving drought tolerance.

ERA1, a gene identified in Arabidopsis, encodes the ß-subunit of a farnesyl-transferase, and is involved in ABA signaling. Plants lacking *ERA1* activity have increased tolerance to drought, however are also severely compromised in yield. In order to have a conditional, reversible down-regulation of ABA, a group of Canadian researchers used a drought-inducible promoter to drive the antisense expression of ERA1, in both Arabidopsis and canola plants.[5] Transgenic plants performed significantly better under water stress, with consistently higher yields over conventional varieties. Importantly, there was no difference in performance between transgenic and controls in

conditions of sufficient water, demonstrating that the technology has no yield-drag.[5] Multi-location trials have confirmed yield increases due to enhanced protection to drought to be 15-25 percent compared to non-transgenic controls.

ABA-independent gene regulation to drought stress

The transcription factors *DREB1* and *DREB2*, are important in the ABA-independent drought tolerant pathways, that induce the expression of stress response genes. Over-expression of the native form of *DREB1*, and of a constitutively active form of *DREB2*, increases the tolerance of transgenic Arabidopsis plants to drought, high salinity and cold. Although these genes were initially identified in Arabidopsis plants, their presence and role in stress tolerance have been reported in many other important crops, such as rice, tomato, barley, canola, maize, soybean, rye, wheat and maize, indicating that this is a conserved, universal stress defense mechanism in plants.[4] This functional conservation makes the *DREB* genes important targets for crop improvement for drought tolerance through genetic engineering.

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Coping with Salt Stress through Biotechnology

Salinity and agriculture

High salinity in agricultural fields has been a serious problem, because the evaporation of irrigated water of poor quality leaves behind salt solutes which accumulate in the soil over time. While irrigation has made it possible to extend agriculture to semi-arid and arid areas of land, and has been partly responsible for the large increases in food production of the last 40 years, it has also resulted in large-scale water lodging and salinity.

Land degradation due to increased salinity presently affects about 20% of world's area under irrigation, without taking into account arid areas or deserts, which comprise a quarter of the total land of the planet.[1] Most crops are very sensitive to salt, which severely affects yield; increases the severity of other stresses; and can be lethal to the plant. High salt concentration also affects the soil structure, porosity and water retention properties. The development of crop varieties with increased tolerance to abiotic stresses such as drought and salinity is therefore an important strategy to this end.[2]

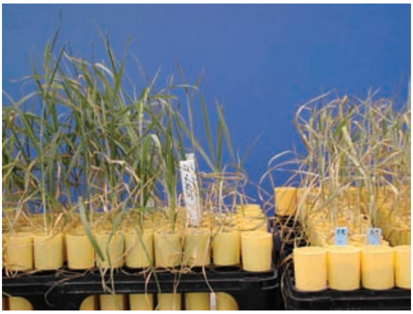
Salt stress

Salt stress effectively decreases the availability of water in the soil to plants, and hence there is a substantial overlap between plant responses to drought and to salinity.[3] Generally, varieties developed to be more tolerant to drought and that use water more efficiently, will also be more resilient to salt stress.[4,5] However, in addition to affecting the water balance of the plant, salt poses another problem to plants: excess

accumulation of salt ions in cells is toxic and fatal. Salt ions impair enzyme function, inhibit protein synthesis, affect the structure and permeability of cell membranes, inhibit photosynthesis, and lead to the production of toxic reactive oxygen species.

Breeding salt tolerant crops

The existence of plants that thrive in soils with high level of salts (termed halophytes), and the occurrence of variation between crop cultivars in salt sensitivity, indicate that salt tolerance is to a large extent under genetic control. Halophytes represent only about 2% of plant species, however, they can be found among half of the terrestrial plant families and are very variable and diverse. Although the development of tolerance to salt is believed to have occurred independently several times during the evolution of land plants, halophytes seem to have evolved the same basic method for dealing with salinity: storing harmful salt ions in the cell vacuole and accumulating organic solutes (which act as osmoprotectants) in the cell cytoplasm.[6]



Exposure to high salinity is killing Yecoro Rojo (right), a wheat cultivar that has moderate salt tolerance. Plants resulting from hybrid crosses with W4910 (left) show much greater tolerance of high salinity.

Conventional breeding requires the identification of genetic variability to salinity among different varieties or cultivars of a crop, or in sexually compatible species, and breeding this tolerance into lines with suitable agronomic characteristics. Genomic tools, such as molecular markers and gene profiling methods, can greatly improve the efficiency of breeding programs.

Engineering salt tolerant crops

The genetic control to salt stress differs in different stages of the plant's life cycle: tolerance at the adult stage does not necessarily correlate with tolerance at the seedling and juvenile stages, or to the ability to germinate in the presence of salts.[2,7]

Tissue-cultured *Arabidopsis thaliana*

Mutant analysis — the screening for mutations that affect the plant's response to stress — has been a crucial tool in the discovery of genes acting in the network. Screens designed include those aimed to identify mutations with increased or decreased sensitivity to drought, salinity and cold stresses. Also important has been the use of DNA microarray technology, which allows monitoring changes in gene expression in response to stress, and to identify genes that are either induced or repressed by the treatment.[5]

The development of salt tolerant crops by genetic engineering have focused on the following strategies: increasing the plant's ability to limit the uptake of salt ions from the soil; increasing the active extrusion rate of salt ions; and improving the compartmentalization of salt ions in the cell vacuole where they do not affect cellular functions. Genes encoding osmoprotectants have also been the targets of genetic modification experiments, but although their over-expression in some cases improves salt tolerance, in general they also affect plant growth in the absence of stress with negative effects in yield, a highly undesirable trait for farmers.[2,7]

Salt intake is controlled by low and high affinity ion transporters: transmembrane proteins that move ions across the cell membrane, which are also required for the intake of potassium ions (K+). The efflux of ions from the plant depends on the activity of the *SOS1* gene (for *Salt Overly Sensitive1*), initially characterized in Arabidopsis but recently identified in rice, and shown to be functionally conserved between dicots and monocots.[8] Vacuolar membrane transporters, including the one encoded by *AtNHX1* gene of *Arabidopsis*, play a role in the sequestration of ions into the vacuole. NHX1 proteins are also conserved across species, and have been isolated from several crops. Over-expression of *NHX1* genes in Arabidopsis, rice, canola and tomato have been reported to increase the tolerance to salt stress.[3]

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Biotechnology and Biofortification

The ideal diet: sufficient and balanced

A major challenge of our time is that one sixth of the world's population suffers from hunger, a situation which is totally unacceptable. In addition, over half of the global population are afflicted by hidden hunger, which is due to the quality, rather than the quantity, of the food available.[1]

Nutrient deficiencies pertain mainly to proteins and micronutrients. Dietary supplements and food fortification programs are used to address this problem. However, such programs have limitations: target populations are often not reached; often not sustainable over time; address mostly the symptoms and not the cause of the problem. Introducing biofortified staple crops can therefore have a very big impact, as the strategy relies on improving an already existing food supply.[2]

Biotechnology and biofortification

Biofortification capitalizes on the consistent daily intake of food staples, thus indirectly targeting low-income households. Biofortified seeds are also likely to have an indirect impact in agriculture, as a higher trace mineral content in seeds confers better protection against pests, diseases, and environmental stresses, thereby increasing yield.[3]

Biofortified crops can be developed by traditional breeding methods, provided there is sufficient genetic variation in crop populations for the desired trait. In staple grains like rice, improvement of some complex traits such as vitamin A is not possible using conventional breeding, as there are no natural rice varieties rich in this vitamin. All plants produce pro-vitamin A, but only in the green plant parts and not in the starch-storing part of the seed. Conventional breeding is also very difficult in vegetatively propagated varieties (such as cassava and potatoes), due to the scarcity of genetically well-defined breeding lines. In addition, conventional breeding cannot change important traits of the crops desired by consumers, such as taste. Genetic engineering represents therefore a very valuable, complementary strategy for the development of biofortified crops.

Increased protein content

Human cells can produce only 10 out of the 20 amino acids, the building blocks of proteins, and so the missing essential amino acids must be supplied in the food daily. In many poor developing countries, the daily intake of essential amino acids is often not sufficient due to the scarcity of high-protein sources like meat, fish, or soybean.

Suitable protein candidates for biofortification include the storage protein Sporamin A from sweet potato, the seed albumin AmA1 protein from Prince's Feather (*Amaranthus hypochondriacus*), and ASP1, an artificial storage protein rich in essential amino acids. ASP1 has been introduced and expressed successfully in rice and cassava, and efforts are under way to optimize expression and increase the level of protein accumulation in transgenic plants.

Combating Vitamin A deficiency

Vitamin A deficiency causes irreversible blindness, and increased susceptibility to disease and mortality. Rice plants produce β-carotene (provitamin A) in green tissues, but not in the seeds. A public-private partnership to produce rice varieties rich in provitamin A culminated in the development of Golden Rice, in which two genes were introduced by genetic engineering. These encode the enzymes phytoene synthase (PSY) and phytoene desaturase (CRTI). Golden Rice 1 contains the PSY gene from daffodil and the CRTI gene from the bacterium *Erwinia uredovora*, both expressed only in the rice seed.[4] Replacing PSY with genes from maize and rice increased the level of β-carotene by 23 times in Golden Rice 2.[5] Half the daily recommended allowance of vitamin A for a 1-3 year old child would therefore be provided for in 72g of Golden Rice 2. Golden Rice is in advanced testing stages.

Iron-rich crops against anemia
Iron deficiency anemia affects more than 2 billion people in virtually all countries, which makes iron deficiency by far the most common micronutrient deficiency worldwide. Iron is found in vegetables, grains, and red meat. However, the bioavailability of iron in plants is low, and in rice, the problem is aggravated by the presence of phytate, a potent inhibitor of iron resorption, and by the lack of iron resorption-enhancing factors.

Therefore, scientists had to increase the iron content in grains, reduce the level of phytate, and add resorption-enhancing factors.[6] Expression of the iron storage protein ferritin from French bean and soybean in the endosperm of rice results in a 3-fold increase of iron in seeds. In order to decrease the level of phytate, an enzyme that degrades it (known as phytase) has also been transformed into rice, and efforts are currently under way to optimize the construct. Finally, overexpression of a cysteine-rich protein that transports metals in rice can improve the rate of iron resorption during digestion.

Increased folic acid in tomato
Folic acid deficiency is a global health problem that affects mainly, though not exclusively, women in developing countries.

In food, most of the folic acid occurs as folate. In order to engineer high folate tomatoes, scientists overexpressed the genes encoding the enzymes catalyzing the synthesis of two folate precursors.[7] This resulted to transgenic tomato fruits with up to 25 times more folate than controls.

Phytase maize
In China, phytase maize has been developed by Chinese Academy of Agricultural Science and was given a biosafety certificate in 2009, and renewed in 2014.[8]

Challenges for biotech biofortified crops

A major problem of developing biofortified crops is the cost of research and of regulatory compliance, due to the extreme precautionary regulation of biotech crops. In the case of biofortified crops, where profit margins for private technology developers are slim, the scarcity of public funds exacerbates this problem.[9] GM technology tends to be proprietary, so intellectual property issues also need to be duly considered.

A successful biofortification strategy requires widespread adoption of the crops by farmers and consumers, and this presents several important challenges.[9] Public acceptance is essential, especially if the new trait changes perceptibly the qualities of the crop, such as color, taste, and dry matter content. Wide dissemination of the technology also relies on good market networks and channels for the dissemination of agricultural information.

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Applications/Products

Contributions of Agricultural Biotechnology in Alleviation of Poverty and Hunger

Reduction of poverty and hunger are key priorities and targets of top global agenda. However, the number is increasing at the rate of four million a year, with Africa having the largest proportion of people living in absolute poverty. Agriculture remains predominantly traditional and majority of countries in the region exhibit a high dependency on food aid, which accounts for a quarter of all global food aid shipments. Reversing this trend requires strategic interventions that would dramatically raise agricultural productivity while taking into consideration realities and diversity of Africa's farming systems.

Stark reality of hunger and poverty status

- Poverty causes more sickness, suffering and death than any other problem on earth
- One fifth of humanity are afflicted by a vicious cycle of poverty
- Most people worst hit by hunger and poverty are in developing countries
- Majority of the countries perpetually experiencing food emergencies globally are in Africa

Trends in modern biotechnology

The global area under biotech crops has increased at unprecedented rate since the first year of adoption in 1996. With this, the contribution of biotech crops to reducing poverty is significant. Furthermore, the global net economic benefit to biotech crop farmers in developing countries and the reduction in environmental footprints have been significant.

Which way out?

Agriculture accounts for 70% of fulltime employment, 33% of total GDP and 40% of total export earnings in Africa. Yet, productivity level of most crops fall below global averages.

- At the onset, African farmers face a multitude of highly complex and interrelated problems
- No single approach will provide solutions to the declining agricultural productivity trends
- Conventional crop improvement alone will not cause a dramatic "quantum jump" to bridge the huge food deficit and poverty face of Africa

Agriculture accounts for 70% of fulltime employment, 33% of total GDP and 40% of total export earnings in Africa. Yet, productivity level of most crops fall below global averages.

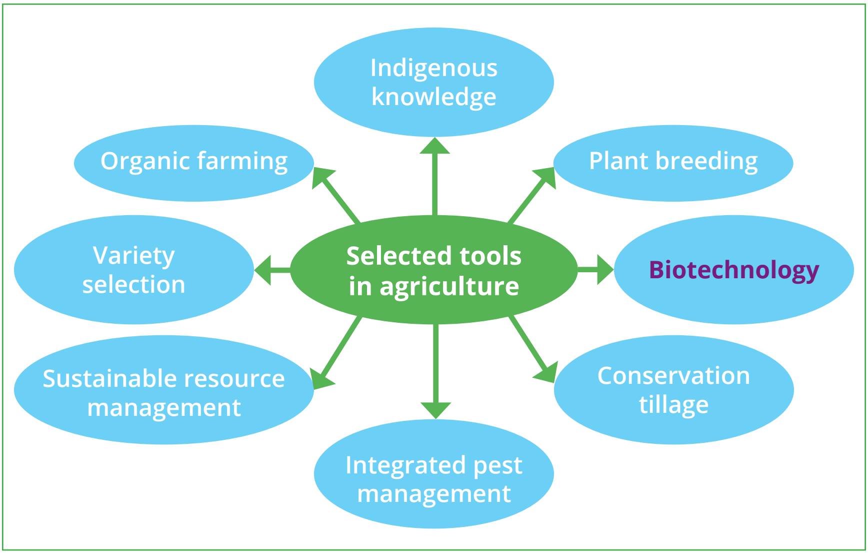

Figure 1. Several tools are used to increase agricultural productivity

The case for modern agricultural biotechnology

Biotechnology enables diverse applications in agriculture, health, industry, and the environment. Overwhelming evidence demonstrates that biotechnological tools — tissue culture, genetic engineering and molecular breeding (marker-assisted selection) continue to provide promising opportunities for achieving greater food security while improving the quality of life. Biotechnology however is not a magical bullet. A high quality seed requires good agronomic practices, appropriate inputs and support services for the farmer to reap benefits. The comparative advantage of currently available biotech crops is the built-in defense against insects and tolerance to weed killers making them suitable for the average farmer. The technology is scale neutral and with proper stewardship, even the very small farmers benefit.

Safety of biotech crops

With over a decade of production and consumption, biotech food and feed products depict a history of safe use with no credible evidence of risks to human health or the environment. This has been confirmed by a number of reputable independent scientific bodies such as the Research Directorate General of the European Union, the French Academies of Sciences and Medicine, and the British Medical Association.

Food and Agriculture Organization (FAO) of the UN has reported: "to date, no verifiable untoward toxic or nutritionally deleterious effects resulting from the consumption of foods derived from genetically modified foods have been discovered anywhere in the world."

Health benefits of biotech crops

Besides reduction in pesticide residues, biotech crops have potential to increase the nutritional value of foods and enhance human health in various ways:

- Lower levels of infestation by insects reduces fungal and mycotoxin in maize.

- Nutritionally enhanced rice for beta-carotene, would provide an alternative source of vitamin A to save millions of children who go blind every year.
- Biotech processes can reduce presence of toxic compounds - e.g. cyanide in cassava.

Environmental benefits of biotech crops

- Cumulative reduction in pesticides has contributed to less pesticide residue in foods and minimized impact on non-target organisms.
- Increased productivity per unit of land, minimizing encroachment into marginal lands, destruction of forests and pollution of fresh water resources.

Moving into the future

Responsible and safe deployment of modern biotechnology can significantly enhance prospects for alleviating poverty and hunger in Africa. To realize the technology's potential however, African governments should create an enabling policy environment and institutional arrangements for investment in R&D and commercialization of these products. Mechanisms to facilitate access to proprietary technologies and to invigorate the public sector towards development of products relevant to local conditions should be strengthened.

One of the major constraints to adoption and utilization of modern biotechnology in Africa is misinformation. This continues to influence acceptance and policy choices. Generation of accurate and science-based information is therefore crucial for informed decision making, which would lead to greater appreciation of the contributions of biotechnology to food security and wealth creation.

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Biotech Rice

Rice is the staple food for two billion people living in Asia and Africa. With the imminent doubling of the world population in 2050, rice production should be increased.[1] Hence, numerous initiatives and strategies were developed towards increasing rice production and helping rice farmers in the developing world.

Development of biotech rice started in the early 80's. This period overlapped with the development of different genetic engineering procedures for rice.

Pest and disease-resistant biotech rice

With the discovery of resistance genes, breeders have aimed to improve rice's resistance to pests. Stemborer resistance breeding has been difficult since there is no high level of resistance in the rice gene pool. However, laboratories have developed different local varieties containing Bt genes, individually or in combination, for resistance against lepidopterans.[2,3,4]

Bacterial blight (BB), caused by *Xanthomonas oryzae* pv. *Oryzae* causes huge yield losses. The discovery of the *Xa21* gene, which confers broad-spectrum BB resistance, started the development of rice varieties with the *Xa21* gene.[5]

Development of rice resistance to sheath blight were also conducted by incorporating genes coding for enzymes that metabolize the pathogenic-related proteins.[6,7] Herbicide tolerant rice has also been developed. Rice resistant to glufosinate, a natural, contact herbicide that controls a wide range of weeds, was a novel weed control measure in 1999.

Abiotic stress resistance

Rice uses 30% of the freshwater used for crops worldwide.[8] With the imminent water shortage and increased salinity brought by climate change, development of resistance to abiotic stresses gained prominence.

Stress-related genes and transcription factors were identified in Arabidopsis and used in rice. The *HRD* gene increased the rice leaf biomass and probably enhanced photosynthesis and drought resistance.[9] *CBF3/DREB1A* and *ABF3* genes were also expressed in rice, increasing its salinity and drought tolerance.[10] Bacterial genes for trehalose accumulation also increased tolerance to drought, salt, and cold in transgenic rice.[11]

Nutritional improvement

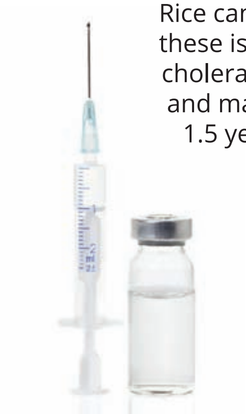
Rice is a good source of carbohydrate, proteins, fiber, lipid and fats, minerals and vitamins.[12] In poor countries where rice is predominantly eaten, important minerals and vitamins are lacking in the diet. This leads to a widespread vitamin A and E, iron and zinc deficiency, affecting children, and pregnant and lactating women. Food supplementation and fortification programs conducted were expensive and ineffective.

A novel approach is biofortification, which uses biotech tools to increase amounts of essential nutrients. Golden Rice has been developed[13,14] and is being used to transfer beta carotene into high-yielding cultivars in the Philippines, Bangladesh and India. Rice with increased ferritin content were also developed to solve global iron deficiency.[15]

Ordinary rice (left) and golden rice (right) (Source: www.goldenrice.org)

Plant proteins are primary sources of proteins in the diet and improving their quality, such as done in biotech rice with improved lysine content, will be significant in the future.

Biopharming in rice



Rice can also be used to produce pharmaceuticals. One of these is the rice-based oral vaccine containing the antigen cholera toxin B subunit (CTB). The vaccine remained stable and maintained efficacy at room temperature for more than 1.5 years. Other vaccines can be produced in rice to target diseases and can be administrated economically in the developing countries.

Extended use of antibiotics usually leads to the development of antibiotic resistance in commensal bacteria in poultry, pigs, cattle, and humans, prompting the search for alternative strategies. Antibacterial molecules such as lactoferrin and lysozyme were considered and expressed in rice grains through biotechnology. Broiler chickens fed with rice containing lactoferrin and lysozyme showed improved feed efficiency and intestinal health. This strategy can also be used in maintaining intestinal health in young animals including humans.[16]

Biotech rice and the future

Biotech rice has been developed to address concerns on rice production such as pests, diseases, and abiotic stresses. However, it has also addressed nutrition, environmental safety, and global warming. Studies to increase rice yield are underway, such as the use of the C4 pathway, in order to convert light energy and carbon dioxide into assimilates more efficiently.[17] Research on apomictic rice or the production of cloned seed has also been started.[18] This will reduce the cost of production of hybrid rice.

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Biotech Wheat

Wheat is a staple food processed into flour and used for different types of breads, pastries, pastas, and cereals. It is also used for fermentation of alcoholic beverages and biofuels. *Triticum aestivum* (bread wheat) and *Triticum durum* (durum wheat) are the commonly grown species today.

Wheat is the second most-produced cereal crop after maize. Maize and soybean are getting ahead of wheat in terms of production because conventional efforts for wheat are not keeping pace with the modernized techniques used to improve maize and soybean. Thus, there is renewed emphasis on utilizing modern biotechnology to produce more and better wheat.

Herbicide tolerant wheat

In 2007, BASF released Clearfield, the first herbicide tolerant wheat developed through mutation breeding to survive the presence of imidazolinone herbicide. Based on the results of the field trials in the U.S., Clearfield is almost similar to the parental line in terms of vigor, time to maturity, yield, disease resistance, and tendency to weediness.[1]

The first herbicide tolerant wheat produced through genetic engineering was MON71800 (Roundup Ready™ wheat). A gene from common soil bacterium was introduced to wheat to produce a glyphosate tolerant wheat line. The gene codes for the production of a novel form of the enzyme 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS) which functions in the biochemical synthesis of compounds vital for growth and survival. Although studies have proven that MON71800 is safe and nutritious just like the other conventional wheat varieties[2], the developers decided not to introduce the GM wheat variety to the market.

The future biotech wheat

Pest resistant wheat

Wheat is affected by fungal diseases such as stem rust (*Puccinia graminis*), *Septoria*, *Fusarium* and common bunt (*Tilletia tritici*) which can easily spread in the fields when the surrounding area is moist. Among these fungal pests, *Fusarium* is the worst, causing crown rot and head blight that result to production of small and stunted grains or no grain at all. Some *Fusarium* strains also produce mycotoxins, which are harmful for humans and animals. Syngenta has been working on Fusarium-resistant wheat but postponed the project in 2007 due to public concern over biotechnology. This could be a candidate for reconsideration with the growing interest for biotech wheat.[3] Syngenta also partnered with International Maize and Wheat Improvement Center (CIMMYT) to develop stem rust resistant varieties of wheat through marker-assisted breeding.[4]

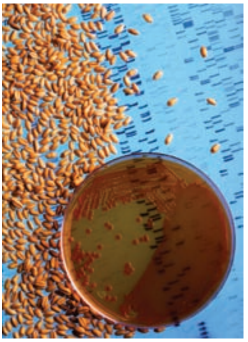
Healthy wheat head (left) in contrast to the one inoculated with *Fusarium graminearum*, showing severe symptoms of scab

In 2009, researchers from the Institute of Plant Pathology in Zurich and John Innes Center in Great Britain separately revealed two rust-resistance wheat genes that could be the best solution in eliminating the rust fungus threat.[5] The *Lr34* gene isolated by Zurich researchers could be responsible in fighting off diseases. John Innes Center scientists identified the *Yr36* gene in wild wheat, which has a potential to provide resistance to modern wheat varieties.

The Chinese Academy of Agricultural Sciences (CAAS) possibly has the highest investment in the world for GM wheat. They are developing several traits in wheat such as resistance to yellow mosaic virus, head scab, powdery mildew, and insects. The Henan Agricultural University is also developing sprouting-tolerant wheat, to get rid of the 20% loss in production due to early sprouting.[6,7]

Salt tolerant wheat

CSIRO Plant Industry researchers have isolated two salt tolerance genes (*Nax1* and *Nax2*), which came from the old wheat relative *Triticum* *monococcum*. Both genes inhibit sodium, which can be toxic to plants, by limiting its passage from the roots to the shoots.[8] Based on the field trials, the lines with the *Nax2* gene produced 25% more yield than those without the gene in saline conditions.



Biofortified wheat

Wheat is also being developed to be safe for people with celiac disease, which is caused by the consumption of gluten that leads to damage to the small intestine. Washington State University (WSU) is conducting experiments using genetic techniques to remove the celiac-causing gliadins in the wheat grain with improved baking quality traits. The variety is also expected to contain more lysine.[9]

Drought tolerant wheat

In 2007, 30 wheat transgenic lines were tested in Victoria by the Department of Primary Industries project. Each wheat line contains six different drought tolerance genes from maize, Arabidopsis, moss, and yeast. These genes encode proteins that will regulate biochemical pathways to promote normal growth under reduced amounts of water.[10] Similarly, CIMMYT used a gene (*DREB1A*) from Arabidopsis to enhance the characteristics of wheat. The GM wheat exhibited tolerance to drought, low temperature, and salinity.[11]

The second chance of biotech wheat

The acceptance for biotech wheat has changed over the years. A 2009 wheat growers survey conducted by the National Association of Wheat Growers showed that 76% of the respondents are in favor of the petition supporting the commercialization of biotech wheat.[12] International Food Information Council also reported that 73% of their consumer respondents said they would likely purchase bread, crackers, cookies, cereal, or pasta made with GM wheat developed to use less water, land and/or pesticides.[13] Nine wheat-related associations from Australia, Canada and the U.S. released a GM Wheat

Trilateral Statement, announcing the need for more investment in R&D of GM wheat.[14]

The International Wheat Genome Sequencing Consortium produced a survey sequence, covering 61% of bread wheat's genome, together with in-depth of the cereal's largest chromosome. This will help scientists develop better wheat varieties.[15]

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Biotechnology for Sugarcane

Sugarcane, from the genus *Saccharum*, accounts for 80% of sucrose produced worldwide. Around two billion metric tons of sugarcane stalks are crushed in sugar mills annually. But there is certainly more to this crop than sugar.

With traditional technologies, sugarcane can yield a variety of products from fiber to chemicals. With modern biotechnology, this crop can be grown and used in more diverse ways. Plant genetic engineering promises to turn sugarcane into a more efficient producer not only of sucrose but also of biofuels and compounds with medical and industrial uses.

Boosting the sucrose yield

Genetic manipulation is being used to increase sucrose content of sugarcane. This requires understanding the interacting processes involved in accumulation of sucrose in stems. Scientists have identified the key enzymes that start these processes, which can be hastened or slowed down by genetic engineering to more efficiently build-up of sucrose in stems.[1]

In sugarcane, genetic modification is done one step at a time to boost the sucrose yield. For instance, the first step is when South African scientists genetically knocked down a particular enzyme, increasing the sucrose in young stems of the modified plants.[2] This indicates the potential for substantial improvement in sucrose yield of sugarcane through precise modification of underlying processes.

Making cellulosic biofuel

Sucrose is widely used for making ethanol. Ethanol is an alternative to fossil fuel, which can reduce greenhouse gas emission. Breeders have focused on sucrose yield to boost ethanol production. However, the increased use of sucrose for ethanol has raised ethical and economic concerns. These stressed the need to produce ethanol without sacrificing sucrose.

Biotechnology seeks to tap the cellulose in leaves and bagasse, the residue from crushed stalks, for ethanol production. The chemical structure of cellulose can be degraded by enzymes into simple sugars and can be fermented into ethanol. However, the presence of a tough material called lignin makes the procedure relatively costly.

Genetic engineering in Brazil aimed to modify lignin so that it can be easily separated from the bagasse, allowing a more efficient conversion to ethanol.[3] In Australia, researchers inserted microbial genes into sugarcane, creating plants that can make cellulose-degrading enzymes.[4] Both studies could impact the cellulosic ethanol technology.

Biofactory for niche products

Scientists find sugarcane as an ideal plant for the co-production of substances for medical and industrial applications. The genetic mechanisms within sugarcane cells can be tweaked to use them to produce these substances, turning it into a biofactory. Engineered sugarcane plants were shown to produce high-value chemicals and natural precursors of biopolymers.[5,6,7]

A breakthrough in this area is the production of an alternative sweetener, isomaltulose, in transgenic sugarcane. This was achieved by inserting a bacterial gene for an enzyme that transforms sucrose into isomaltulose.[8] As a sweetener, isomaltulose may bring certain health benefits as it is good for diabetics, and it does not support the growth of bacteria that cause tooth decay.

Enhancing crop productivity

Genetic engineering may boost the productivity of sugarcane to a higher level for farmers and to accomplish other objectives. Genes from other organisms can also be inserted into sugarcane to protect it from harsh environmental conditions and pests. The first transgenic sugarcane which will be released in Indonesia, contains a bacterial gene for drought tolerance.[9]

Transgenic approaches have also been used to control insect pests, diseases and weeds that limit the productivity of sugarcane. The introduction of a gene from a soil bacterium gives sugarcane resistance to stemborers.[10] Infection of sugarcane by a harmful virus can be prevented by inserting a gene derived from the virus itself.[11] A bacterial gene responsible for detoxification of a certain class of herbicide has conferred an attractive trait for weed control.[12]

Key challenges

The potential of the sugarcane biofactory has drawn scientific and business interests, but its commercial use would be a huge regulatory challenge, as it is intended for field cultivation. Thus, the commercial viability of a sugarcane biofactory will depend on the efficiency of risk containment.

Sugarcane researchers are optimistic about the impact of the transgenic sugarcane, arguing that potential benefits far outweigh the risks. This must be effectively communicated to address the negative perception of consumers and traders toward transgenic crops and their products.

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Bt Eggplant

Eggplant is a staple food in India and other countries in South and Southeast Asia where it is called ***brinjal***, along with over 30 Sanskrit names.[1] It also comes in a variety of shapes and colors. Some are striped and round, others white and short, looking like a chicken egg, thus its most famous name.

In the Philippines, eggplant is known as ***talong*** and is the number one vegetable in terms of production area. An average of 21,225 hectares is planted with eggplant each year.[2] In India, it is grown on nearly 550,000 hectares, making the country the second largest producer after China with a 26% world production share.[3] In Bangladesh, it is the third most important vegetable in terms of production and grown on about 50,000 hectares across the country.[4] Hence, eggplant is an important source of income to many Asian farmers.

The benefits of eggplant do not stop with the farmers. It is also beneficial for human health because it is high in fiber and water, rich in anti-oxidants, and a good source of vitamins and minerals.[5] This vegetable can help prevent cancer, diabetes, and gastrointestinal diseases.

Eggplant growers' pest problem

Eggplant farmers suffer significant yield losses at 51-73% annually due to the Fruit and Shoot Borer (FSB).[6] Female moths deposit eggs mostly on eggplant leaves. When the eggs hatch and turn into larvae (Figure 2), they feed on leaf tissues and tunnel inside shoots and fruits (Figure 3).

Fig. 1. Adult moth (Rao, 2010)

To address this problem, many eggplant farmers in major eggplant producing areas in the Philippines and Bangladesh spray chemical insecticides every other day, or up to 80 times per growing season.[7,8] The practice is unacceptable

Fig. 2. FSB larvae

and unhealthy to consumers, farmers, and the environment. It is also a common practice in the Philippines to dip unharvested eggplant fruits in a mix of chemicals to ensure marketability of fruits.[7] In India, farmers spray insecticides 20-40 times per crop cycle or else they will have no harvest.[9]

Bt technology for eggplant

Fig. 3. Non-Bt eggplant

Fig. 4. Bt eggplant

(Photos: UPLB IPB Eggplant Project, 2014)

Bt stands for *Bacillus thuringiensis*, a common soil bacterium that contains a gene which produces a protein harmful to FSB. Scientists have incorporated this gene to eggplant to confer insect resistance.

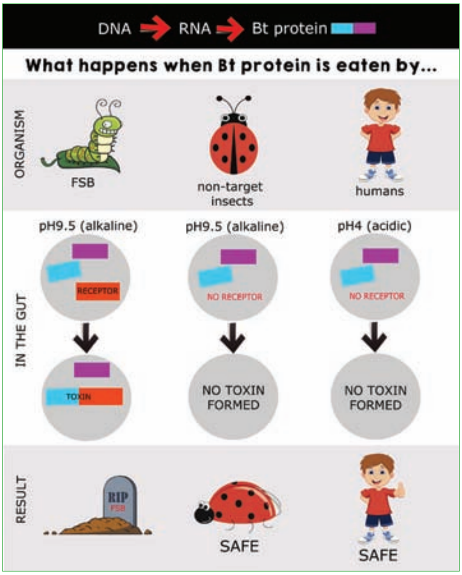

Fig. 5. Fate of Bt protein in FSB, non-target organisms, and humans

Bt eggplant (Figure 4) expresses Bt gene, enabling it to produce the same protein that makes it resistant to FSB. When Bt protein is ingested by FSB larva, it is made soluble by the presence of enzyme and alkaline condition (pH9.5) of the gut. It then binds into another protein (receptor) present in the midgut resulting to an active toxin. The Bt toxin then punctures the gut leaving the insect unable to eat. The insect dies within a few days.[7] The Bt protein only affects FSB and does not affect humans, farm animals, and other non-target organisms because these organisms do not have the required gut conditions (pH and required receptor) to activate the toxin (Figure 5).

Other Bt crops such as Bt corn and Bt cotton have showed improved pest management and reduced insecticide use. This also led to greater net return for growers and improved conditions for non-target organisms.

Commercialization of Bt eggplant

Bangladesh

Bangladesh is the first country in the world to allow the commercial planting of Bt brinjal when it approved four varieties in October 2013 in time for the 2013-2014 growing season.[10] The government agencies approved the following varieties of Bt brinjal: Bt Uttara, Bt Kajla, Bt Nayantara, and Bt ISD006. In January 2014, Agriculture Minister Matia Chowdhury distributed Bt brinjal seedlings to 20 Bangla farmers.[11] Based on experimental data, Bt brinjal can increase yield by at least 30% and reduce the frequency and cost of insecticide applications by 71-90%.[4]

India

In India, Bt brinjal was developed by the Maharashtra Hybrid Seeds Company (Mahyco).[3] Despite the conduct of field trials from 2002-2006, a moratorium was issued in October 2009, and a government ban was implemented in February 2010.[12] Results of the multilocation research trials showed that Bt brinjal can reduce insecticide use by 77%. Marketable fruits may increase to 116% over conventional hybrids and 166% over popular open-pollinated varieties. Researchers have estimated that Bt brinjal will deliver farmers a net economic benefit of Rs.16, 299 (US$330) to Rs.19,744 (US$397) per acre with national benefits to India exceeding $400 million per year.[3]

Philippines

Studies were conducted on the potential costs and benefits of Bt eggplant commercialization in the Philippines based on the results of multi-location field trials of the crop. The average potential net benefit of planting Bt crop is PhP272,000 (US$6,243)/ha higher than conventional varieties in the province of Pangasinan and PhP120,000 (US$2,753) in Camarines Sur. This significant increase in profit is due to increased marketable yield and reduced pesticide use. It was projected that there will be 48% reduction in pesticide application per hectare. This can be translated to 19.5% lower environmental footprint compared to non-adopters.

Aside from the increase in income, significant health and environmental benefits will accrue from the considerable reduction of pesticide use.

Assuming a 50% adoption rate, the benefits of Bt eggplant to human health is valued at PhP2.5 million (US$57,353) per year while the collective benefits to farm animals, beneficial insects and bird is estimated to be PhP6.8 million (US$155,841) per year.[2]

The Philippine Court of Appeals (CA), however, issued a Writ of Kalikasan to stop the field trials of Bt eggplant in the country on September 20, 2013.[10] The respondents have since filed a petition to the Supreme Court to review the CA decision.

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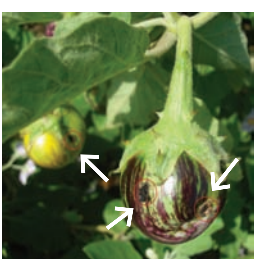
Bt Brinjal in India

Brinjal or baingan, known as eggplant and aubergine in other parts of the globe, is a very important vegetable in India. Low in calories and high in nutrition, the vegetable has high water content and is a good source of fiber, calcium, phosphorus, folate, and vitamins B and C. It is also used in ayurvedic medicine for curing diabetes, hypertension, and obesity. Dried brinjal shoots are used as fuel in rural areas.

Brinjal is grown on nearly 550,000 hectares in India, making the country the second largest producer after China with a 26% world production share. It is an important cash crop for more than 1.4 million small, marginal and resource-poor farmers. Brinjal, being a hardy crop that yields well even under drought conditions, is grown in almost all parts of the country.

In spite of its popularity among farmers, brinjal cultivation is often input intensive. Brinjal is prone to insect attack, the most destructive of which is the fruit and shoot borer (FSB) *Leucinodes orbonalis*. FSB larvae bore into tender shoots and fruits, retarding growth, making the fruits unsuitable for the market and unfit for human consumption. Fruit damage as high as 95% and losses of up to 70% in commercial plantings have been reported.



FSB-damaged fruits

Farmers resort to frequent insecticide applications and biological control measures to counter the threat of FSB. However, since FSB larvae are concealed within shoots and fruits, the pest normally escapes insecticide sprays. Therefore farmers tend to over-spray insecticides, which lead to negative effects on the environment, high pesticide residues in vegetables and serious risk to consumers' health and safety.

Several attempts have been made to develop resistant cultivars through traditional plant breeding, but these have met with limited success. Thus, scientists used biotechnology to develop FSB-resistant brinjal variety.

India's first vegetable biotech crop

FSB-resistant brinjal or Bt brinjal was developed using a transformation process similar to the development of Bt cotton. Bt brinjal incorporates the *cry1Ac* gene expressing insecticidal protein to confer resistance against FSB. The *cry1Ac* gene is from the soil bacterium *Bacillus thuringiensis* (Bt). When ingested by the FSB larvae, the Bt protein is activated in the insect's alkaline gut and binds to the gut wall, which breaks down, allowing the Bt spores to invade the insect's body cavity. The FSB larvae die a few days later.

Bt brinjal was developed by the Maharashtra Hybrid Seeds Company (Mahyco). The company used a DNA construct containing the *cry1Ac* gene, a CaMV 35S promoter and the selectable marker genes *nptII* and *aad*, to transform young cotyledons of brinjal plants. A single copy elite event, named EE-1, was selected and introduced into hybrid brinjal in Mahyco's breeding program. Mahyco also generously donated the Bt brinjal technology to the Tamil Nadu Agricultural University (TNAU), Coimbatore and University of Agricultural Sciences (UAS), Dharwad. The event EE-1 was backcrossed into open-pollinated brinjal varieties.

The National Center on Plant Biotechnology (NRCPB) and Indian Institute of Horticultural Research (IIHR) have also developed varieties expressing *cryFa1* and *cry1Ab* genes, respectively.

Biosafety and food safety assessments

Rigorous scientific tests, including toxicity and allergenicity evaluation as well as nutritional studies on rabbits, rats, carps, goats, broiler chickens and dairy cows, have confirmed that Bt brinjal is as safe as its non-Bt counterparts. The safety of Bt brinjal was further validated by the results of the studies on pollen escape, effects on soil microflora and non-target organisms, agronomy, invasiveness and Bt protein degradation. Results of the studies demonstrated that Bt brinjal does not affect beneficial insects.

Climbing the regulatory ladder

Since its development in 2000, Bt brinjal has undergone rigorous scientific evaluation to assess its safety.

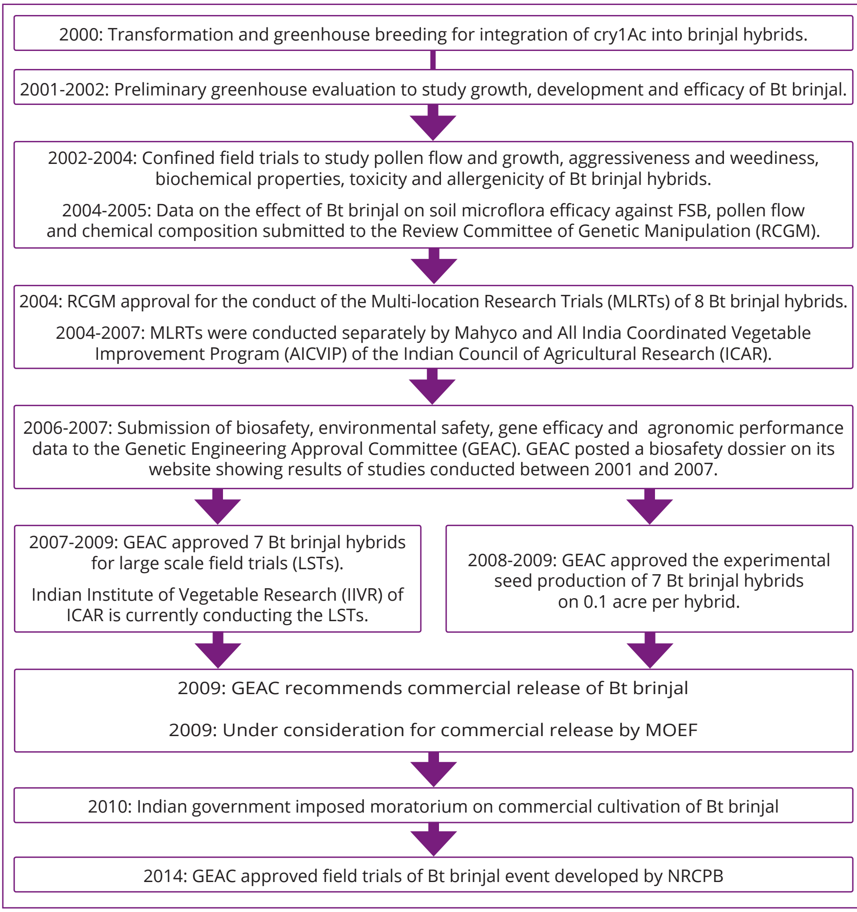


Figure 1. Development and regulation of Bt Brinjal in India

Farmer and consumer benefits

Bt brinjal was found to be effective against FSB, with 98% insect mortality in Bt brinjal shoots and 100% in fruits compared to less than 30% mortality in non-Bt counterparts. The multilocation research trials (MLRTs) confirmed that Bt brinjal required 77% less insecticides than non-Bt counterparts for control of FSB, and 42% less for the control of all insect pests of brinjal. The benefits of Bt brinjal, translate to an average increase of 116% in marketable fruits over conventional hybrids, and 166% increase over popular open-pollinated varieties (OPVs). Furthermore, the significant decrease in insecticide usage reduced the farmers' exposure to insecticides and results in a substantial decline in pesticide residues in brinjal fruits. Scientists have estimated that Bt brinjal will deliver farmers a net economic benefit ranging from Rs.16,299 (US$330) to Rs.19,744 (US$397) per acre with national benefits to India exceeding $400 million per year.

Conclusion

Studies confirmed that Bt brinjal provide effective control against FSB, and decrease insecticide input by as much as 80%. Bt brinjal also yields significantly more marketable fruit than conventional hybrids and open-pollinated varieties.

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Biotechnology in Ornamental Plants

Ornamental plants are grown for decoration, and aesthetics rather than for food or raw materials.[1] Ornamentals plants are classified into several groups. Cutflowers are fresh cut from plants and are used in flower arrangements.[2] Ornamental grasses are grown fully and used in landscaping as perennials.[3] Lawn or turf grasses are perennial grasses that cover the ground as desired.[4]

Potted and indoor plants are grown indoors for decoration.[4] Bedding plants are grown in greenhouses and are intended to be transplanted to a flower garden.

Trees and shrubs are used for landscaping. Trees include cherry blossoms and palms while Hibiscus and Ficus species are the common shrubs.[4]

Uses of biotechnology in ornamental horticulture

Cellular engineering

Micropropagation. This is used for plant multiplication of elite varieties of ornamentals. It is suitable for ornamentals as it is faster and creates exact replicas.[5]

Haploid breeding. Using this method, haploid individuals, with only half the number of chromosomes, are obtained resulting in dwarfed plants.[5]

Polyploid breeding. Polyploids, with multiple sets of chromosomes, are used for their improved characters such as larger flowers.[5]

Genetic modification

Genetic modification (GM) is also used in the field of ornamental plants because development of new varieties via hybridization or mutagenesis is very difficult.[6,7] Several traits of ornamental plants have already been modified.[8] Some ornamental traits are more valuable to the growers while others have more value to consumers. Some traits are for breeders such as traits that affect seed production.[9]

Table 1. Genes used in the development of GM ornamentals

Gene and Source(s)	Result(s)	Reference(s)
***F3'-5'h* gene** *Petunia/Pansy*	overexpression produces blue flowers in combination with a silenced *dfr* gene in Carnation (Petunia) and Roses (Pansy)	Katsumoto et al. 2007
CrtW *Lotus japonicus*	overexpression changes petal color from light yellow to deep yellow or orange in Lotus	Suzuki et al., 2007
CHS *Gentian*	gene silencing produces white flowers in Gentian	Nishihara et al., 2006
ANS *Gentian*	gene silencing produces pale blue flowers in Gentian	Nakatsuka et al., 2008
Ls *Chrysanthemum*	less branching in Chrysanthemum	Han et al., 2007 Jiang et al., 2009
Ipt *Agrobacterium tumefaciens*	increased branching and reduced internode length in Chrysanthemum	Khodakovskaya et al., 2009
RolC *Agrobacterium rhizogenes*	dwarfed *Pelargoniums* and *Petunias*	Boase et al.,2004; Winefield et al., 1999
MADS-Box Orchid/Lily	ectopic expression changes the second round of petals into calyx in orchids and lilies	Thiruvengadam and Yang, 2009
Asl38/lbd41 *Arabidopsis*	flowers turned into multiple column patterns in *Celosia cristata*	Meng et al., 2009
Floral integrator genes *Arabidopsis*	activate the floral identity genes; promotes flowering in appropriate conditions	Amasino and Michaels, 2010; Jung and Muller, 2009; Turck et al., 2008
AP1 Chrysanthemum	speeds up time to flowering in Chrysanthemum	Jiang et al., 2010; Shulga et al., 2010
Cry1A *Bacillus thuringiensis*	resistance to *Helicoverpa armigera* and *Spodoptera litura* in Chrysanthemum	Shinoyama and Mochizuki, 2006; Soh et al., 2009
CVB coat protein gene Chrysanthemum	Chrysanthemum Virus B (CVB) resistance	Skachkova et al., 2006
Rdr1 Rose	resistance to black spot in Roses	Kaufmann et al., 2003
Sarcotoxin gene *Sarcofaga peregrina*	resistance against *Burkholderia caryophylli* in Carnation	Yoshimura et al., 2007

Table 1. Genes used in the development of GM ornamentals (continued)

Gene and Source(s)	Result(s)	Reference(s)
Rd29A:DREB1A *Arabidopsis*	enhanced abiotic stress tolerance in Chrysanthemum	Hong et al., 2009; Hong et al., 2006a; Hong et al., 2006b
ACO/ACS-coding genes carnation/apple	increased vase life in carnation	Inokuma et al., 2008; Veres et al., 2004
ERS1 chrysanthemum	mutated gene slows down yellowing of leaves in Chrysanthemum	Narumi et al., 2005
Cp4 Epsps *Agrobacterium tumefaciens*	Glyphosate herbicide tolerance in creeping bentgrass	Chai et al., 2003

Major genetically modified ornamentals

Carnation

The "Moon" series, with various flower colors, from Suntory Limited and Florigene are the only GM products commercialized and have been available since the 1990s.[10,11]

Chrysanthemum

Molecular breeding for the blue chrysanthemum is still on-going.[12,13] However, other traits of chrysanthemum are also being improved.[6,13,14]

Roses

Suntory released the famous "blue" rose, "APPLAUSE", in Japan in 2009 and in North America in 2011. Researchers in Suntory are now using different approaches, including other bluing factors to develop a true blue rose.[15]

Petunia

The petunia-CHS, with altered flower colors, from Beijing University is the only event commercially available.[10] Ornamental Biosciences in Germany is now focusing on improved abiotic stress resistance, specifically frost tolerance.

Future prospects

It can be expected that more GM ornamentals will be released in the future.[5,7] Moreover, ornamental horticulture may be compatible with the production of pharmaceuticals and would be a promising approach.[7]

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Nitrogen Use Efficient Biotech Crops

Nitrogen is one of primary macronutrients important for plant growth and development, particularly in metabolic processes such as production of nucleic acids, proteins, and other helper molecules. It is a basic component of chlorophyll, which is vital for photosynthesis.[1] Nitrogen is abundant in the atmosphere but is not readily available for plant use. It can only be used up by plants when it is converted into ammonia from fixation by bacteria.

Nitrogen use efficient biotech crops

Biological nitrogen fixation occurs in some plants with the help of bacteria. One common symbiotic bacterium involved in nitrogen fixation is known as *Rhizobium* which attacks and reproduces in the roots of legumes to get their nutrition. After about a week of infection, white or grey nodules form in the roots. The bacteria through the action of the enzyme nitrogenase, convert the nitrogen gas into ammonia. The plants use ammonia to produce amino acids and other nitrogen-containing molecules.[2] Then the nodules increase in size and turn pink, indicating that nitrogen fixation has occurred.[3] Plants that do not form associations with bacteria must get nitrogen from the soil. However, frequent use of soils in farmlands becomes depleted with nitrogen. Thus, nitrogen fertilizers are applied.

Use of synthetic nitrogen has increased dramatically leading to significant boost in crop yields. However, only 30-50% of the applied nitrogen is absorbed by the plants and the wasted nitrogen has considerable impacts on the environment. It can contribute to algal bloom and reduced oxygen in water leading to loss of aquatic life; and depletion of the ozone and global warming. Thus, scientists seek for more environment-friendly and cost-effective strategies to improve nitrogen use efficiency of crops. One of these strategies is the use of biotechnology.[2]

Genetic engineering nitrogen use

Improving the nitrogen use efficiency of plants requires manipulation of several genes. Genes from different sources have been found to be involved in the process and were investigated if their modification can lead to improved nitrogen use of plants (Table 1). [4]

Table 1. Genes studied for improvement of nitrogen use

Gene(s)/source	Results	Reference
nif **genes** *Klebsiella pneumoniae*	activated nitrogenase function in *Escherichia coli*	Swain and Abhijita, 2013
GS1 tobacco	enhanced grain yield and biomass; improved nitrogen content in wheat, tobacco, and maize	Oliveira et al., 2002
AS1 *Arabidopsis*	improved soluble seed protein content, total protein content, and better growth in nitrogen-limiting medium	Lam et al., 2003
Dof1 maize	improved growth under nitrogen limiting conditions; enhanced nitrogen assimilation	Yanagisawa, 2000
OsNADH-GOGAT1 rice	increase in spikelet weight of up to 80 percent in rice	Yamaya et al., 2002
AlaAT barley	production and degradation of alanine (functions as an intercellular nitrogen and carbon shuttle) in rice	Shrawat et al., 2008
STP13 *Arabidopsis*	improved plant growth and nitrogen use	Schofield et al., 2009

Development of nitrogen use efficient (NUE) crops

Corn

In 2008, DuPont and Arcadia Biosciences announced that they completed five years of multiple field trials of corn which resulted to improved NUE and thus can lead to improvement in farm economics as well as environmentally positive effects.[5]

Wheat

In 2012, Australian Centre for Plant Functional Genomics and Commonwealth Scientific and Industrial Research Organisation (CSIRO) announced their collaboration with Vilmorin & Cie in developing NUE wheat with the aim of reducing nitrogen fertilizer use in Australia.

Developing NUE wheat will significantly impact 35% of the world population where wheat is a staple crop and represents 20% of the total protein intake.[6]

CSIRO has applied for a license for dealings involving 17 wheat lines and 10 lines of barley which have improved nutrient utilization efficiency on a limited scale and under controlled conditions.[4]

Rice

Arcadia Biosciences, African Agricultural Technology Foundation, and the International Center for Tropical Agriculture reported that in 2013, two years of field trials of NUE rice was completed in Colombia. The researchers integrated the NUE technology with New Rice for Africa (NERICA) varieties developed by Africa Rice Center. Results showed that transgenic rice lines out-yielded the conventional NERICA variety by 22 percent on the first year of trial and 30 percent by the following year.[7]

Canola

As of 2007, Arcadia Biosciences has completed five seasons of field trials of canola. The results of the trials showed that the canola plants had the same yield as the conventional varieties, but only half of the required nitrogen input was used. When the same amount of nitrogen with the conventional plants was used, the yield increased by about 15 percent.[8]

Sugarbeet

SES VanderHave and Arcadia Biosciences have conducted three years of field trials to assess the yield performance of NUE sugarbeet varieties. Experimental varieties produce higher yields than controls under various fertilizer applications over multiple years. They are now preparing regulatory data which will become available for all NUE technology licensees.[9]

Sugarcane

South African Sugarcane Research Institute and Arcadia Biosciences announced in 2011 their collaboration in producing high-yielding sugarcane varieties that requires half the amount of the nitrogen fertilizer needed by conventional sugarcane varieties.[10]

Future outlook on nitrogen use

A long-term tracer study revealed that 30 years after application of nitrogen fertilizer to agricultural soils in 1982, 12–15% of the fertilizer-derived nitrogen was still residing in the soil organic matter, while 8–12% of the fertilizer had already leaked toward the groundwater. Part of the remaining nitrogen fertilizer present in the soil is predicted to continue to be taken up by crops and to leak toward the groundwater in the form of nitrate for at least another 50 years, much longer than previously perceived.[11] With the development of NUE crops, such environmental concerns would be dispelled or reduced. Farmers would also lessen economic losses for nitrogen fertilizer, and use their resources for other farm inputs to get more harvest.

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Functional Foods and Biotechnology

Functional foods provide health benefits aside from basic nutrition. These foods contain antioxidants that may lower the risks from certain diseases associated with aging. Examples of functional foods include fruits and vegetables, whole grains, soy, milk, enhanced foods and beverages and some dietary supplements.

Diet and health are closely related. Thus, crops are now being enhanced through biotechnology to increase levels of important biologically active substances for improved nutrition and to remove undesirable food components.

Higher levels of phytosterols for reduced cholesterol

Phytosterols and phytostanols are cholesterol-like molecules found in all plant foods, but the highest concentrations occur in unrefined plant oils, including vegetable, nut and olive oils. Nuts, seeds, whole grains, and legumes are also good dietary sources of phytosterols.[1] Studies have shown that these compounds can lower the risk of cardiovascular diseases.

As phytostanols are more stable than phytosterols during food processing, genetic engineering has been applied for the development of rapeseed and soybean oils with modified ratios of phytosterols to phytostanols.[2] Plants were transformed with a gene from yeast encoding the enzyme 3-hydroxysteroid oxidase, which converts phytosterols to phytostanols.

Higher levels of carotenoids for increased Vitamin A

Carotenoids are yellow, orange, and red pigments found in plants. Some carotenoids are converted by the body into vitamin A, which is essential for normal growth and development, immune system function, and vision.[1] Examples of carotenoids present in plants include α- and

β-carotene (carrots and pumpkins), lycopene (tomatoes) and lutein and zeaxanthin (dark green leafy vegetables).

Transgenic plants that have been developed with increased carotenoid production include:

- β-carotene fortified rice (Golden Rice)
- Canola with increased carotenoids[3]
- Tomatoes with increased β-carotene[4,5]

Higher levels of antioxidants

Pollution, radiation, cigarette smoke, and herbicides generate harmful free radicals in our body, which can cause damage to the DNA and proteins, harm cellular components, and can eventually cause cancer.

Antioxidants are important biological compounds that can protect the body by neutralizing the activity of free radicals. Antioxidants occur in different forms, phenolic compounds such as flavonoids and tocopherols being the most common. They are found in most fruits and vegetables such as carrots, broccoli, and berries.

To enhance the flavonoid content of potatoes, Lukaszewicz and colleagues conducted single and multiple-gene transformations for the enzymes in the biosynthesis of flavonoids.[6] Transgenic plants exhibited significantly increased levels of phenolics, and improved antioxidant capacity.

Higher levels of essential fatty acids

Essential fatty acids, “good fats” include linoleic acid (LA), alpha-linolenic acid (ALA) and other polyunsaturated fatty acids (PUFAs). These fatty acids are considered essential because they cannot be synthesized by our body. A large number of scientific studies suggest that higher dietary essential fatty acid intakes are associated with reductions in cardiovascular disease risk.[7]

The main food sources of the long-chain omega-3 fatty acids are fish. Plants lack the enzymes to make long-chain fatty acids needed by mammals.[5] Scientists at the University of Bristol modified Arabidopsis to produce long-chain PUFAs. The transgenic plants were modified with 3 genes encoding different enzymes that convert linoleic and alpha-linolenic acids to the long-chain PUFAs.[5]

Other biotech functional foods

Low linolenic soybean

Oil from soybean seeds contains the unstable linoleic and linolenic acids, which affect its stability and result in the production of harmful fatty components during processing.

Genotypes with elevated oleic acid content and reduced linoleic and linolenic acid levels are therefore desirable to improve the functionality of soybean oil by increasing oil utility at higher temperatures,[8] and by extending its shelf-life. Monsanto developed the VISTIVE™ soybean, which contains less than 3% linolenic acid, compared to 8% for traditional soybeans.

GM soybean lines were produced by silencing a gene that controls the activity of an enzyme responsible for the conversion of linolenic acid from oleic acid.[9] The result is a more heat stable soybean oil which may be used in frying and other food applications.

High lysine maize

Corn has low-lysine and low-tryptophan content in its major seed storage proteins, zeins. Lysine is an important component of animal feeds, especially for swine and poultry. Kernels with reduced levels of zein proteins have been shown to have increased levels of lysine and tryptophan.[10]

A high lysine and high tryptophan transgenic maize was developed by inserting gene constructs that reduced formation and accumulation of α-zeins. In addition, a large increase of accumulated free amino acids (asparagine, aspartate and glutamate) was observed in the zein-reduced kernels.[10]

Opportunities and challenges

Staple starchy crops have been modified to reduce amylopectin, which is associated with diet-related conditions such as diabetes. In areas of drought and poor soil quality, where high quality proteins are scarce, genetic modification has been used on some legumes and in soybean to increase the levels of high quality proteins.[11]

Commercialization of GM nutritionally-enhanced crop is very limited due to many factors that include the cost of introducing a new product to the market and the lack of suitable regulatory controls.

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Nutritionally-Enhanced GM Feed Crops

Animal industry's demand for feeds is expected to grow as need for food increases simultaneously with human population. Sufficient and nutritious feeds are essential in order to provide food for the population. Nutrition enhancement in crops aims to increase desirable components as well as decrease the undesirable ones through metabolic engineering.

Feed crops with improved proteins and amino acids

Biotechnology has been successfully used in developing crops with increased amino acids, providing an alternative to food supplements. An example would be GM maize with increased lysine, which was found to be as safe and nutritious as conventional maize.[1]

Other examples of these include the protein-enriched soybean event M703 and a narrow-leafed lupin (*Lupinus angustifolius*) with increased methionine content.[2]

Feed crops with biologically active substances

Some crops were modified to contain active substances to increase the nutrition of feeds. Scientists expressed a *Bacillus* ß-glucanase in barley to make it a possible alternative to maize in areas where maize is unavailable.[3] Researchers also introduced human lactoferrin (LF) and lysozyme (LZ) genes in rice grains as a substitute for antibiotics in poultry diets.[4]

Feed crops with improved phosphorous availability

Phosphorus (P) is not thoroughly used in the digestive tract of monogastric animals without the enzyme phytase.[5] Hence, a GM corn with the *Escherichia coli*-derived phytase gene was developed. The GM corn was found as effective as the commercial phytase.[6]

Several other phytase genes were also expressed in other crops. These include GM soybean tobacco and wheat with *Aspergillus niger* phytase transgene[5] as well as GM canola with phytase gene from *Aspergillus fucuum*.[7]

Feed crops with reduced toxins and anti-nutritive factors

Non-ruminants are affected by anti-nutritive factors in plant tissues. A combination of genetic engineering and conventional plant breeding led to the reduction or removal of the major anti nutritive factors in crops for animal feeds.

Soybeans contain raffinose and stachyose, oligosacharides that cause osmotic problems in laboratory animals. Hence, genetically modified soybeans with low oligosaccharides were developed. These soybeans have lower levels of anti-nutritive factors as well as a higher crude protein and sucrose contents.[8]

Cottonseed meal has been used in cattle feeds for improving growth and breeding. However, it contains gossypol which can cause reduced performance. Researchers reduced the gossypol in cottonseed through genetic modification. The resulting cotton had low gossypol content and maintained the crop's ability to resist insect pests.[9]

Gaps in nutrition enhancement of GM feed crops

Nutritionally-enhanced GM feeds have shown efficacy in providing nutrients to poultry and livestock. Sufficient and cheap feeds are expected to come as more countries are adopting biotech crops. Research on improving other feed components such as vitamins, minerals, fats, as well as reducing anti-nutrition factors in plant-based feeds, and efficient anaerobic fermentation of silage will surely be looked forward to in the coming years.

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Molecular Pharming and Biopharmaceuticals

With genetic engineering, scientists are able to modify living organisms to produce specific pharmaceuticals. Biopharmaceuticals are drug products produced in living systems and used for therapeutic or diagnostic purposes or as dietary supplements.

The use of plants to express proteins is more practical, safe and economical compared to other systems. Plants allow low-cost production since expensive equipment are not required. The production of these compounds in plants is called molecular pharming.

The first protein expressed in plants was the human serum albumin, produced in 1990 in transgenic tobacco and potato. Years after, two plant-derived pharmaceuticals (PDPs) or plant-made pharmaceuticals (PMPs) have been commercialized. There are also various veterinary applications of plant-derived vaccines and therapeutic proteins. Table 1 shows examples of plant-derived pharmaceuticals for human diseases that are in the research pipeline.

What plants are used for biopharma production?

Transgenic tobacco is the most popular choice in studies on plant-produced proteins due to its high biomass yields and is not used as food or feed.[1] It has recently been used in Cuba for the commercial production of a recombinant antibody against hepatitis B2. Potato was the first to be used for vaccine production.

Leafy crops have also been studied for biopharmaceutical production. Harvested material from leafy crops must be processed immediately due to its short shelf life. To solve this, cereal grains are being developed to produce the PDPs.[1] Maize has already been used for the commercial production of recombinant avidin, *b*-glucuronidase and trypsin.[2]

Table 1. Plant-derived pharmaceuticals for the treatment of human diseases that are in the pipeline for commercialization

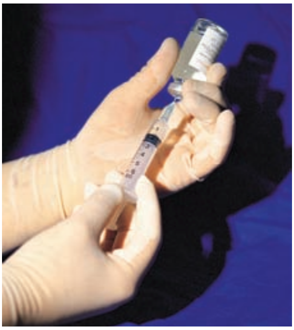
Product	Class	Indication	Crop
Various single-chain Fv antibody fragments	Antibody	Non-Hodgkin's lymphoma	Viral vectors in tobacco
CaroRx	Antibody	Dental caries	Transgenic tobacco
E. coli heat-labile toxin	Vaccine	Diarrhea	Transgenic maize Transgenic potato
Gastric lipase	Therapeutic enzyme	Cystic fibrosis, pancreatitis	Transgenic maize
Hepatitis B virus surface antigen	Vaccine	Hepatitis B	Transgenic potato Transgenic lettuce
Human intrinsic factor	Dietary	Vitamin B12 deficiency	Transgenic *Arabidopsis*
Lactoferrin	Dietary	Gastrointestinal infection	Transgenic maize
Norwalk virus capsid protein	Vaccine	Norwalk virus infection	Transgenic potato
Rabies glycoprotein	Vaccine	Rabies	Viral vectors in spinach
Cyanoverin-N	Microbicide	HIV	Transgenic tobacco
Insulin	Hormone	Diabetes	Transgenic safflower
Lysozyme, Lactoferrin, Human serum albumin	Dietary	Diarrhea	Transgenic rice

Source: Compiled by ISAAA, 2007.

How would biopharma benefit developing countries?

Agriculture is important to developing countries and economies would benefit from a plant-based pharmaceutical platform. PDPs present a cost-effective method of pharmaceuticals that could help control diseases in developing countries.

With plants, production of biopharmaceuticals can be modified to suit local production. Bringing the technology closer to the target population would result in greater involvement from countries, and shift the focus of



current drug production to specific regional diseases.[3] This would alleviate problems associated with the delivery of conventional vaccines and medicines to remote areas.

Demand for specific pharmaceuticals is very high especially in developing countries. The use of PDPs would undoubtedly assist vaccination programs in developing countries by reducing the costs of vaccine production, purification, storage and administration.[3]

What are the risks, concerns, and issues of PDPs?

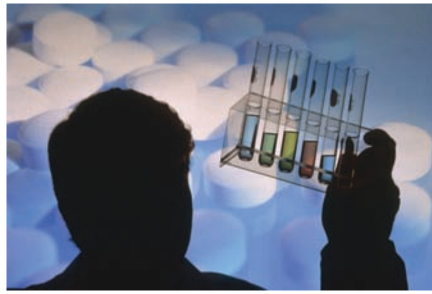
The production of plant-derived pharmaceuticals introduces several unique challenges for biosafety regulation since plants are grown in an open environment. An important concern is therefore the potential gene flow through pollination or seed contamination. In addition, there are issues about PDPs accidentally entering the food chain and being consumed by non-target organisms.

It is impossible to keep the environmental risks associated with PDPs at absolute zero.[4] A realistic approach would be to minimize the environmental exposure of these proteins through a combination of precautionary measures including physical isolation, the use of genetic use restriction technologies, or the expression of the proteins in a form that must be activated.[5]

A major concern for developing countries is the lack of biosafety framework for GM plants. Without a biosafety framework, developing countries cannot perform trials of PDPs[1]. Adoption of PDPs may also raise ethical and religious issues such as the idea that altering living organisms is like ‘playing God’.[6]

Conclusion

The production of plant-derived pharmaceuticals may provide a cheaper and alternative source of medicines for both developed and developing countries. The latter will benefit more from PDPs due to reduced costs of drug production and the complementarity of the technology with agriculture.



Locally grown crops can be developed for PDPs, which could make them more economical for use in developing countries. However, there are risks, concerns, and other issues which need to be addressed before this technology is commercially released.

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Biotechnology for the Livestock Industry

Meat and milk from farm animals including livestock and poultry are sources of high quality protein and other nutrients.[1] Hence, meat consumption has grown with increase in population. However, poor countries may not be able to sustain the daily requirement for meat and milk, leading to malnutrition. Moreover, demand for meat and milk production is also expected to double in 2050 in developing countries.

Thus, increasing production and the safe processing and marketing of meat and milk, and their products are big challenges for producers. Biotechnology is being harnessed in aspects of the industry to hasten breed development for improved animal health, reproduction, and nutritional quality and safety of animal-based foods.

Reproductive animal biotechnology

Various biotechnology methods are used in improving the breeding stock of animals. These include artificial insemination (AI), embryo transfer (ET), in-vitro fertilization (IVF), somatic cell nuclear transfer, and the somatic cell nuclear transfer.

Artificial insemination. Artificial insemination (AI) is where new breeds of animals are produced through the introduction of the male sperm from one superior male to the female reproductive tract without mating. AI reduces sexually transmitted diseases, lessens the need to maintain breeding males, and facilitates more accurate records of pedigrees.[2]

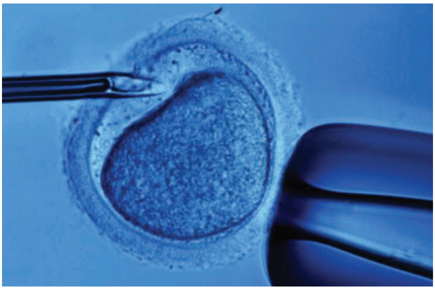

Figure 1. Microscopic view of sperm implantation during in vitro fertilization.

In-vitro fertilization. In case other artificial reproductive techniques fail due to difficulties such as blocked reproductive systems, non-responsive ovaries in the females, low semen quality, and presence of diseases, in vitro fertilization (IVF) is used. The fertilization of the sperm and the egg is conducted in vitro at specific conditions (Figure 1). Successful IVFs have been conducted in various animal species, including humans.[3]

Embryo transfer. Embryo transfer (ET) from one mother to a surrogate mother makes it possible to produce several progenies from a superior female. Selected females are induced to superovulate and inseminated. Embryos are flushed out of the donor's uterus, isolated, and inserted into the uterus of surrogate mothers (Figure 2).

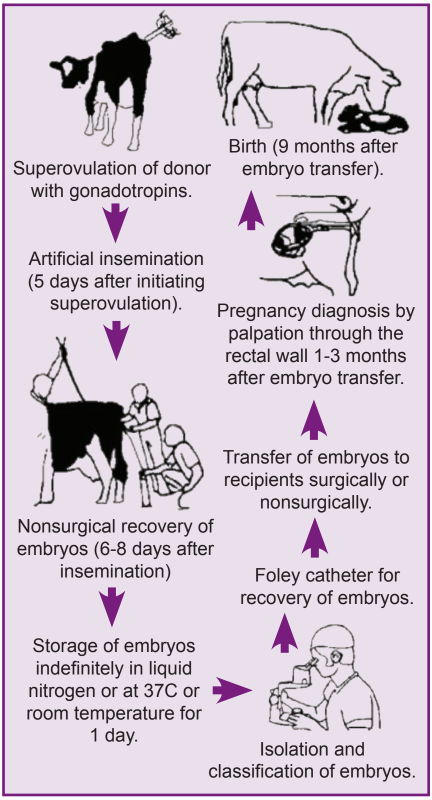

Figure 2. Bovine embryo transfer procedures. (http://cruachan.com.au/embryo_transfer.htm)

ET increases reproductive rate of selected females, reduces disease transfer, and facilitates the development of genetic stocks as well as the production of closely related and genetically similar individuals that are important in livestock breeding.

Somatic cell nuclear transfer. (NF) This is a technique where the nucleus of a somatic cell is transferred into a female egg cell with no nucleus to generate genetically identical individuals.[4] This technique was used to generate Dolly from an adult mammary cell.[2] NF creates the possibility of generating clones of superior genotype and can be used to evaluate effects of genotype x environment interactions. However, high rate of pregnancy loss as well as other problems makes this a pre-commercial technology.

Genomics and marker-assisted selection (MAS) applications

The discovery of DNA sequences or molecular markers associated with important animal traits has various applications including trait improvement, heritability determination, and product traceability.

Molecular marker-assisted introgression (MAI)

Markers are used to guide livestock breeders in selecting individuals expressing the gene. With the use of molecular markers, backcrossing cycles incurred in selection and identification of the desired individual are reduced. Today, molecular markers are being used in various livestock trait improvement activities.[5]

Parentage, product traceability and genotype verification

Molecular markers are tools used by regulatory bodies to ensure product quality and food safety. Livestock parentage and its products can be identified using molecular markers from farm to the consumer's plate. A similar DNA-based technology has also been developed to detect the presence of around 211 bp fragments to facilitate testing of very small meat samples from the supermarket.[6]

Screening for undesirable genes. Genetic diseases and physical defects can be traced and documented in livestock animals using molecular markers.[6] The cause and origin of these problems can be easily traced to the genetic changes and DNA mutations as they manifest in the protein structure and function.[7] With DNA testing, animals carrying these defective genes are easily identified and culled from the program.

The future of DNA-based technology in livestock improvement

With advances in sequencing animal genome, the progress in molecular marker technology, and the use of reproductive biotechnology, there will be numerous research opportunities to improve the livestock industry. In the future, it will be possible to obtain information on the genetic constitution of animals that will allow prediction of the potential of an animal at birth, as well as the selection of animals best suited to a specific production environment.

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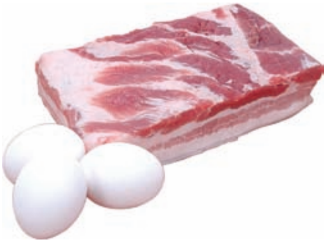
Contribution of GM Technology to the Livestock Sector

GM crops have caused significant benefits to both farmers and consumers. It has minimized the use of pesticides and provided higher crop yields and improved product quality. GM crops have also benefited the livestock sector through increased yields and quality of feed ingredients.

Future demand for livestock products and feed grains



The demand for livestock products will increase dramatically as population increases. Moreover, urbanization and rising income will cause consumption of meat, milk, and eggs to rise.[1] Global demand for meat is also set to increase by 2020.[2] Thus, demand for feed grain will increase. Higher production for food and feed has to come from increased yield due to the limited cultivated area.

GMO materials in GM feed ingredients

Transgenic crops used for feed are modified for herbicide tolerance, insect resistance, oil content, and virus resistance. Proteins expressed in GM crops are safe and are similar to naturally-occurring proteins. Insect resistant crops express proteins from *Bacillus thuringiensis*, a soil bacterium used as insecticide by organic farmers.[3]

GM feed ingredient in livestock diets

Livestock producers in many parts of the world prefer corn grain and soybean meal for energy and/or protein source in both monogastric and ruminant diets.

In 2014, around 38,898 million bushels, or 988 million metric tons, of corn were produced worldwide. Forty percent of the global production was allotted to animal feed. Given that 93% of maize produced in the U.S.

is GM and the U.S. contributing for 36% of world production, then at least 13,219 million bushels or 335 million metric tons of feed maize for 2014 is genetically modified.[4]

Safety assessment of GM products

GM crops pass through extensive testing and a long approval process. This includes comprehensive analyses to ensure food, feed, and environmental safety before being commercialized.

The first step in safety assessment of GM products is to determine if the product is equivalent to conventional counterparts. Further analysis then focuses on the defined differences.

Safety of GM feed crops

Nicolia et al. (2013) analyzed 1,783 studies on safety of GM crops published from 2002 to 2012. The main concerns about GE food/ feed consumption are safety of the inserted genes, the proteins from transgenes, and the change in crop composition. Based on findings, there were no significant hazards linked to GE crops.[5]

Feeding trials have also been conducted to examine the safety of GM feeds for livestocks.[6] These studies reveal no evidence of significantly altered nutritional composition, deleterious effects, or the occurrence of transgenic DNA or protein in products from animals fed with GM feeds.

Table 1. Some GM crops used for livestock feed

Feed Crop	Improved Traits	No. of Approved GM Events
Alfalfa	herbicide tolerance, modified product quality	4
Argentine Canola	herbicide tolerance, modified product quality, pollination control system	32
Bean	viral disease resistance	1
Chicory	herbicide tolerance, pollination control system	3
Cotton	insect resistance, herbicide tolerance	44
Creeping Bentgrass	herbicide tolerance	1

Table 1. Some GM crops used for livestock feed (continued)

Feed Crop	Improved Traits	No. of Approved GM Events
Flax	herbicide tolerance	1
Maize/Corn	modified product quality, insect resistance, herbicide tolerance, pollination control system, abiotic stress tolerance	124
Papaya	disease resistance	2
Plum	disease resistance	1
Polish Canola	herbicide tolerance	4
Potato	insect resistance, disease resistance, herbicide tolerance, modified product quality	40
Rice	insect resistance, herbicide tolerance	4
Soybean	modified product quality, herbicide tolerance, insect resistance, altered growth/yield	27
Squash	disease resistance	2
Sugar beet	herbicide tolerance	3
Tomato	modified product quality, disease resistance, insect resistance	11
Wheat	herbicide tolerance	1

Source: ISAAA GM Approval Database (2015). http://www.isaaa.org/gmapprovaldatabase/

Animals perform in comparable manner when fed GM feed as compared to conventional products. Feeding of GM crops has not shown any negative effects to any farm animal.[6] Studies have also showed that transgenic DNA in GM crops are not detectable in raw food products from GM fed animals.[7,8] Products from GM-fed farm animals are as safe as traditional counterparts.

Future of GM feed crops

GM feed ingredients will continue to benefit livestock with improved feed qualities. Future GM feed crops will also have enhanced nutritional characteristics.[9,10] Researchers are also looking to improve digestibility of some crops. GM crops expressing antigens from microbes are also being developed. Edible vaccines delivered via feeds can potentially control diseases in livestock.

Conclusion

The first generation of GM crops has directly benefited livestock production through increased quantity and quality of feed. Future GM crops with enhanced output traits can improve animal productivity and performance. These will definitely contribute to helping feed the growing world population.

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Biotechnology for Green Energy: Biofuels

Biofuels are alternative fuels made from plant resources used mainly for transportation. There are two types of biofuels: bioethanol, the substitute fuel for petrol for vehicles produced by sugar fermentation of cellulose, and biodiesel, which is produced from oil crops. Examples of crops used for biofuels are shown in Table 1.

Table 1. Main energy crops worldwide

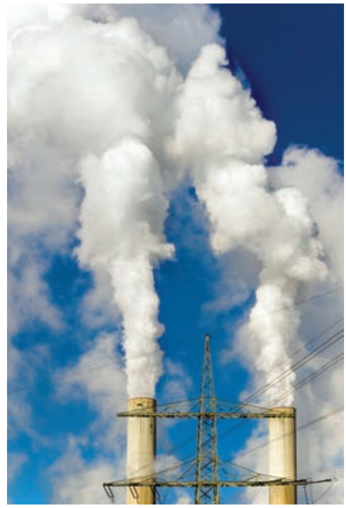
Country	Bioethanol	Biodiesel
Brazil	sugar cane	---
United States	maize	soybean
China	sweet sorghum	rapeseed, sunflowerseed
Germany	sugar beet	rapeseed, sunflowerseed
France	sugar beet	rapeseed, sunflowerseed
Italy	---	rapeseed, sunflowerseed
Canada	cereals	---
Thailand	cassava	---
Spain	sugar beet	---
Denmark	---	rapeseed, sunflowerseed
Czech Republic	---	rapeseed, sunflowerseed
Australia	cereals, sugar cane	---

Sources: United States Department of Agriculture, United States Department of Energy, the European Commission

Why have biofuels?

Meeting future energy demands

Energy demand is projected to grow by 2025. Majority of energy is derived from fossils fuels, which are limited, non-renewable and polluting. Switching to biofuels for transportation needs would reduce dependency on oil and could boost rural development by providing farmers with an additional source of income.



Reducing greenhouse emissions

Greenhouse gases are causing global warming, with fossil fuel and electricity consumption being the main sources. The Kyoto Protocol, an international agreement made under the United Nations Framework Convention on Climate Change (UNFCCC) was established to monitor greenhouse gas concentrations in the atmosphere.

Environmental advantages of biofuels

The main advantage of biofuels is that it is carbon-neutral since the carbon dioxide they release upon combustion was initially extracted from the atmosphere by plants, resulting in zero net greenhouse gas emissions. Biofuels also reduce the release of volatile organic compounds, as mixing ethanol with gasoline burns it more completely. Moreover, biofuels are biodegradable and non-toxic.

The cost of biofuels

The cost of biofuels is estimated not only in terms of energy derived, but also in terms of input required for its production and distribution. The production of energy crops requires land, fertilizers, and machinery, while fermentation and distillation of biofuels need biomass and water. The cost of biofuels will ultimately depend on several parameters that are difficult to quantify such as increased security of supply, effect on climate change, job generation, the impact of an expanding sector on land demand, and its effect on alternative land uses.

Increasing plant yield
Plant growth can be improved by increasing the efficiency of light capture during photosynthesis. The most successful approaches involved introducing genes from photosynthetic bacteria into plants.

Another approach is the manipulation of genes involved in the metabolism of nitrogen, an element in proteins and DNA. Further strategies also include extending the growth phase of plants, to redirect energy for other processes into vegetative growth.

Raising plant protection to abiotic and biotic stress
Pests, pathogens and abiotic stress are the primary causes of crop loss worldwide. Developing crops with improved resistance to stress, pests and pathogens is therefore imperative. For example, transgenic rice overexpressing the glutamine synthase gene (GS2) shows increased tolerance to salinity.

Bt cotton engineered with the insecticidal gene from *Bacillus thuringiensis* is a biotech crop developed with improved resistance to pests.

Optimizing the chemical and physical attributes of biofuel sources
The switch to renewable biomass sources will require the development of energy crops with the desired chemical and physical characteristics.

The feedstock for bioethanol production must shift from grains to low-cost agricultural and municipal wastes. Several approaches would also improve production efficiency from biomass sources such as reducing the proportion of lignin in a plant. An alteration of the properties of the cell wall could also be a strategy for a more efficient release of sugars for fermentation.

Concluding remarks: the road to biofuels

Biofuels can replace transportation energy needs in an environmentally responsible way without affecting global food production.

For biofuels to be instrumental in meeting energy needs, a multidisciplinary approach is required. Biorefinery facilities need to be improved and further research is required to make it sustainable. Commercialization and policy support are also critical.

In addition, socio-economic concerns should be addressed to prevent biofuels from having negative effect on food production or biodiversity.

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Biotechnology for Biodiversity

Biological diversity (biodiversity) is the variability among living organisms: within and between species and ecosystems. Biodiversity is considered as the foundation of agriculture being the source of all crops and livestock species that have been domesticated and bred since the beginning of agriculture.[1] Crops like corn and wheat were inedible wild crops but through years of domestication, edible varieties have been made available.

Biotechnology is presently used for the conservation, evaluation, and utilization of biodiversity particularly for important crops.[2]

Conservation

Extinction and endangerment of species are taking place in different parts of the world, manifesting degradation in the ecosystem.[3] According to the Food and Agriculture Organization, about 75% of the genetic diversity in agricultural crops have been lost over the last century.[4]

The DNA bank is an efficient, simple and long-term method used in conserving genetic resource. Compared to traditional seed or field gene banks, DNA banks lessen the risk of exposing genetic information in natural surroundings. It only requires small sample size for storage and keeps the stable nature of DNA in cold storage. Since whole plants cannot be obtained from DNA, the stored genetic material must be introduced through genetic techniques.[5]

In vitro techniques are also valuable for conserving plant biodiversity.[6] Such techniques involve 3 basic steps: culture initiation, culture maintenance and multiplication, and storage. For medium-term storage, slow growth strategies are applied. For undefined time of storage, cryopreservation is applied, wherein plant tissues turned into artificial seeds and stored at very low temperatures to impede growth. Cryopreservation allows 20% increase in regeneration process compared to other conservation methods.[7]

Evaluating genetic diversity

Germplasm refers to living tissues from which new plants can form. It can be a whole plant, a part of a plant or even just a number of cells. A germplasm holds information on the genetic makeup of the species. Scientists evaluate the diversity of plant germplasms to find ways on how to develop improved varieties.[8] Germplasm evaluation involves screening of germplasm in terms of physical, genetic, economic, biochemical, physiological, pathological, and entomological attributes.[9]

Molecular Markers

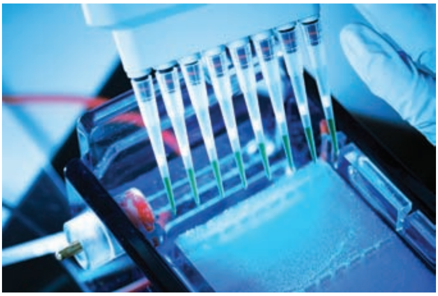
Molecular markers are used to map out the genetic base of crops and select favorable traits to come up with a better germplasm. Molecular markers are short strings of nucleic acid which compose a DNA segment that are closely linked to specific genes in a chromosome. Thus, if the markers are present, then the specific gene of interest is also present.

Marker-assisted selection such as single nucleotide polymorphisms (SNPs), is widely used in different research centers to design genotyping arrays with thousands of markers spread over the entire genome of crops.[10] After observing the desired traits in selected plants, these are then incorporated through modern or conventional breeding methods in existing crop varieties.

Recent advances in genomic, proteomic and metabolomic research offer unique opportunities for the search, identification, and commercial utilization of biological products and molecules in the pharmaceutical, agricultural, and environmental sectors.[11]

DNA and Protein Profiling

To come up with effective conservation management programs for endangered crop varieties, it is important to evaluate their genetic relatedness and distances from other relatives. Such information could be derived through DNA profiling commonly conducted through electrophoresis.



Through this method, an individual organism is identified using unique characteristics of its DNA. DNA profiling depends on sections of the DNA that do not code for a protein. These areas contain repetitive sections of a sequence called short tandem repeats (STRs). Organisms inherit different numbers of repeated sequences from each parents and the variation in the number of repeats within an STR lead to DNA of different lengths. The targeted STR regions on the DNA are multiplied through polymerase chain reaction and then separated by electrophoresis in a genetic analyzer. The analyzer is composed of a gel-filled capillary tube where DNA travels. When electric current is passed through the tube, the DNA fragments move through the gel tube by size. The digital output of the analyzer is read and interpreted through a genotyping software.[12]

The entire set of proteins in a cell is referred to as proteome, and the study concerned with how proteins work and assembled is called proteomics. Proteomics is based on the end-products of gene activity: the protein patterns formed from unique genetic activities. Through 2D acrylamide gel electrophoresis, complex mix of proteins is sorted based on each protein's specific combination of charge and molecular weight. These patterns are standard for protein discovery because the same proteins would migrate at the same points on the gel. The protein bands are developed in digital images and then analyzed in mass spectrometers.[13]

Biodiversity utilization

Most cultivated plant species have lost their inherent traits that came from their wild ancestors. These traits include resistance to harsh environmental conditions, adaptation to various soil and climate conditions, and resistance to pests and pathogens.[8] To utilize these important traits in cultivated varieties, scientists search for the genes that confer such important traits and use conventional (hybridization) and modern biotechnology (particle bombardment or *Agrobacterium*-mediated) to come up with improved genetic variations of crops.

Biodiversity for benefit of all

Biotechnology has raised fears on loss of genetic resources leading to public policy interventions that promote provision of public goods while conserving biodiversity. One of these policies is the N*agoya Protocol on Access to Genetic Resources and the Fair and Equitable Sharing of Benefits Arising from their Utilization to the Convention on Biological Diversity*, which provides a legal framework for the biotechnology industry to manage access to genetic resources and provide fair and equal sharing of benefits.[14]

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Biotech Plants for Bioremediation

Pollutants fall into two main classes: inorganic (heavy metals such as cadmium, mercury, and lead; nonmetallic compounds like arsenic; and radioactive nuclear waste) and organic (solvents, explosives, fertilizers, herbicides, and pesticides).

Conventional remediation for polluted sites typically involves the physical removal of disposal of contaminants. Thus, physical remediation strategies do not eliminate the problem. Such strategies are also expensive, disruptive to the environment, may temporarily increase exposure to chemicals, and often leave residual contamination.

Phytoremediation, the use of plants to remove or degrade contamination from soils and surface waters, has been proposed as a cheap, sustainable, effective, and environmentally friendly alternative to conventional remediation. Plants use solar energy to extract chemicals from the soil and to deposit them in the above-ground part of their bodies, or to convert them to a less toxic form. These plants can then be harvested and treated, removing the pollutants.

Sunflowers extract toxic ingredients from soil, such as lead, arsenic and uranium.

An ideal phytoremediator would have high tolerance to the pollutant; the ability to either degrade or concentrate the contaminant at high levels in the biomass; extensive root systems; the capacity to absorb large amounts of water from the soil; and fast growth rates and high levels of biomass.

Although several species can tolerate and grow in some contaminated sites, these species typically grow very slowly, produce low levels of biomass, and grow in very specific environmental conditions. Trees, which have extensive root systems, high biomass, and low agricultural inputs requirements, tolerate pollutants poorly, and do not accumulate them.

Soil contaminants and their green biotech "mops"

The remedial capacity of plants can be significantly improved by biotechnology. The introduction of novel traits for the uptake and accumulation of pollutants into high biomass plants is proving a successful strategy for the development of improved phytoremediators. Research efforts are underway for efficient bioremediation strategies.

Heavy metals

Toxic metals affect crops and accumulate in the food chain. Metal-tolerant species protect themselves from the toxicity of metal ions by binding metals ions with specific proteins that render them in a safer form. Three classes of proteins are important for metal detoxification in plants: metallothioneins, phytochelatins, and glutathione.

Shrub tobacco (*Nicotiana glauca*) transformed with the phytochelatin TaPCS1 shows high levels of zinc, lead, cadmium, nickel, and boron, and produces high biomass.[1] In Arabidopsis, Indian mustard, and tobacco plants, improved metal tolerance was achieved through the overexpression of enzymes that induce the formation of phytochelatins.[2,3,4]

Plants naturally tolerant to heavy metals have also been used as a source of genes for phytoremediation. Transgenic Arabidopsis plants expressing a selenocysteine methyltransferase (*SMTA*) gene from the selenium hyperaccumulator *Astralagus bisulcatus* contain 8 times more selenium in their biomass when grown on selenite compared to nontransgenic controls. Comparison of gene expression profiles between *Arabidopsis thaliana* and the closely related species *A. hallerri*, which is tolerant to cadmium and hyperaccumulates zinc, is also helping identify major genes required for metal tolerance.[4]

Arsenic occurs naturally in rocks and soil, and is released into underground water. Consumption of contaminated drinking water leads to skin disorders, gangrene, and cancer of the kidneys and bladder. In addition, high levels of arsenic in agricultural land degrade soils, reduce crop yields, and introduce the pollutant to the food chain.[4]

Scientists have engineered Arabidopsis plants with arsenic tolerance by introducing bacterial genes *arsC* and *y-ECS*. *arsC* converts arsenate, the arsenic form absorbed by plants, to arsenite. Double transgenics are not only highly tolerant of arsenic, they also have improved cadmium tolerance, and a six-fold increase in the level of biomass compared to wild-type controls.[5]

Arabidopsis inflorescence

Herbicides

Mammalian P450 cytochrome genes have been used to confer herbicide resistance to crops, which can be used in herbicide rotation systems. These are designed to delay the evolution of herbicide resistance in weeds, and to reduce the environmental load of agricultural chemicals.[3,5] Herbicide resistance is also provided by the overexpression of the maize glutathione S-transferase I (*GSTI*) gene.[5]

Explosives

Explosives, and their degradation products, are extremely toxic and corrosive. Tobacco plants engineered with the bacterial gene for a NADPH-dependent nitroreductase tolerate and degrade high levels of TNT7, and Arabidopsis plants carrying the *xplA* gene from *Rhodococcus* bacteria are highly resistant to Research Department Explosive (RDX), the primary explosive used during World War II.[5]

Landmines

Efforts are underway to develop transgenic plants that can be used to warn civilians of the presence of landmines in a field.[6] Arabidopsis whose roots change color when they come into contact with degradation products of landmines have been developed. Scientists are now working to allow the plant to transmit the signal to their leaves, to effect human-readable changes for a practical explosives detection system.

Mercury

Organic mercury (organomercurials), the most toxic form to living organisms, is produced when bacteria in the water and soil convert elemental mercury into methylmercury. Methylmercury is easily absorbed and accumulates at high levels in the food chain.

Detoxification of organomercurials has been achieved in transgenic plants by transforming Arabidopsis, tobacco, poplar trees, Indian mustard, and eastern cotton wood with two bacterial genes, *merA* and *merB*.[3,5,7] The combined actions of *merA* and *merB* transform methylmercury to the volatile elemental form, which is 100 times less toxic, and is released by the plant to the atmosphere at non-toxic concentrations through transpiration.

Prospects

Although the use of transgenic plants for bioremediation is very promising, several challenges remain such as the need for more understanding on the molecular basis of the pathways involved in the degradation of pollutants; determination of the actual costs of benefits of phytoremediation with biotech plants; and political will and funding support for phytoremediation research especially to implement novel strategies.

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Biotech/GM Trees

Trees provide man's basic needs for food, fuel, shelter, in industry and pharmaceuticals. Trees are known to effectively and efficiently sequester CO2 and other greenhouse gases and slow down the rate of global warming, hence the current global attention on their care and preservation.

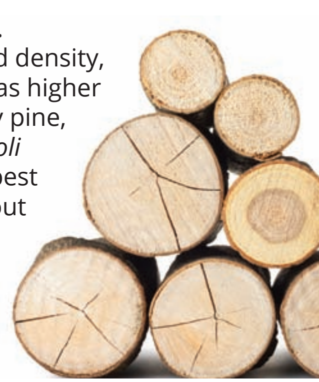
Biotechnology and genetic engineering are tools that can contribute to the improvement of crops and trees. Genetically modified (GM) trees are developed to contain useful traits to lower production costs of wood products, and increase productivity. They improve the economics of tree plantations through creation of suitable raw materials, decreased pesticide use, disease resistance, and rehabilitation of degraded lands. GM trees are useful for the lumber, pulp and paper industry; assure quality and nutritious fruits; and improve forest covers.

GM trees for industrial uses

Trees can be engineered to grow faster, denser, and straighter which are desirable for lumber companies, and can also be used as non food crop-based biofuel feedstocks. This can be achieved by developing trees with more cellulose and less lignin.

FuturaGene has developed GM eucalyptus and poplar trees that contain genes that alter the structure of plant cell walls to stimulate the natural growth process. The GM eucalyptus which was given approval for commercialization in Brazil in 2015[1] can grow 5 meters a year, with 20%-30% more mass at shorter time than normal eucalyptus trees.

Loblolly pines are used for lumber, plywood, and paper. Arborgen has developed a GM cultivar with higher wood density, associated with lumber strength and durability, as well as higher energy content for biomass uses. Genes from Monterey pine, the American sweet gum tree, mouse ear cress and *E. coli* introduced through gene gun were deemed non plant pest risks, thus USDA deregulated the GM loblolly pine without undergoing environmental studies.[2]



Poplar tree was improved to easily break down lignin, which is commonly removed from wood through extensive chemical and energy intensive procedures. Scientists inserted the genes for ferulic acid into the cells creating weak points in the lignin chemical structure. The new lignin can be readily broken for easy processing.[3]

GM trees to combat invasive threats

Engineering trees to make them more resilient to changing climates and are better able to defend against pests and diseases is critical to keep our forests and trees healthy.

Papaya Ringspot Virus resistant papaya

Hawaii in 1997 suffered a devastating 40% economic loss due to papaya ringspot virus (PRSV). The US$17 million papaya industry was saved in 1998 by the U.S. government's Environment Protection Agency (EPA), Food and Drug Administration (FDA), and U.S. Department of Agriculture Animal and Plant Health Inspection Service (USDA APHIS) approval of GM papaya Rainbow resistant to the PRSV disease. Researchers from Cornell University and the University of Hawaii expressed the virus-derived coat protein gene, and within four years of its introduction, papaya production returned to previous level. An additional PRSV Papaya X1' 7-2 was deregulated in the US in 2009.[4] By 2014, GM papaya occupies 1,000 hectares in Hawaii, and 8,475 hectares in China.[5]

Insect resistant poplar

In China, poplar trees are cultivated for use in furniture, boat-making, paper, chopstick and many others because of its flexibility and close wood grain. The emergence of Asian longhorn beetle devastated the 7.04 million hectares of poplar in China. In 2003, China commercialized GM poplar trees that contain *cry1a* genes from *Bacillus thuringiensis* and later with a proteinase inhibitor from *Sagittaria sagittifolia* to control the beetle.[6] In 2014, a total of 543 hectares GM poplar was cultivated in China.[5]

Virus resistant plum

In 2007, USDA deregulated C5 HoneySweet plum tree engineered to contain virus-derived coat protein gene for resistance to plum pox virus. The plum was issued an "approval to use" letter by the US FDA in January 2009 and was conditionally registered by US EPA in August 2011. While not made commercially available as of August 2013, the USDA is posed to make the tree available as needed to combat the virus.[7]

Blight resistant American chestnut

The bacteria *Cryphonectria parasitica* caused the chestnut blight disease that extremely affected the American chestnut forest in the late 19th century. The chestnut blight is caused by oxalic acid released by the pathogen during infection. Researchers at State University of New York (Syracuse) and University of Georgia, expressed a wheat oxalate oxidase in chestnut trees to control the disease. Successive field trials of these transgenic trees showed promising results.[8] Obtaining approvals from USDA, the EPA, and the FDA is expected to take at least five years.

Citrus greening resistant citrus


Citrus greening disease on mandarin oranges (Photo: USDA)

Citrus greening caused by the bacteria *Candidatus liberibacter asiaticus* and spread by psyllids was recorded in the early 70's. The disease turns oranges into green, misshapen, and bitter-tasting fruits. Millions of acres of citrus crops have already been lost in the US and overseas, and 80% of Florida's citrus trees in the US$5.1 billion industry are infected and declining. Cocktails of chemical sprays to kill the vector psyllids are no longer effective. A Texas A&M scientist, with funds from Southern Gardens company inserted a spinach gene to fight the bacteria. Seven-year successive small and larger field trials were completed successfully by 2013. Southern Gardens is now seeking to deregulate these oranges, anticipating first commercial planting in three to four years.[9]

GM tree for adverse temperature

A one of a kind GM eucalyptus tree that can withstand extremely low temperature was developed by Arborgen, Inc. and was deregulated in the USA in 2010. The GM tree contains a cold-inducible promoter driving a C-repeat binding protein from *Arabidopsis thaliana*. Selected transgenic lines were tested in 21 replicated field trials across eight different locations with various freezing temperatures. Transgenic freeze tolerant eucalyptus can grow up to 52.4 feet at 16.8°F, compared to the control trees (0.3 feet).[10]

GM tree for consumers

New generation GM traits are targeted for consumer preference. The "non-browning apple" Arctic® Apple from Okanagan Specialty Fruits of Canada is the first GM tree with consumer-targeted trait which was approved for commercialization in 2015. Polyphenol oxidase (PPO) renders the apple brown upon oxidation when bruised, bitten, or cut. Hence, GM apple was developed by silencing the three main polyphenol oxidase genes making the apple with little or no PPO enzyme, such that cell disruption doesn't lead to browning.[11]

The future

Future GM trees will continue to contain traits for feedstock biofuels, timber industry, to resist pest and diseases, to save contaminated environments (phytoremediation), and with new traits such as drought and salinity tolerance. It can be foreseen that R&D of GM forest trees will develop even faster in the near future and there is no doubt that GM forest trees will be grown on a large scale in plantation forestry and for land reclamation in China, Brazil and the USA as long as some technical obstacles are overcome in the coming years. Regulations in planting GM trees are also needed in many countries to ensure environmental safety before commercialization.

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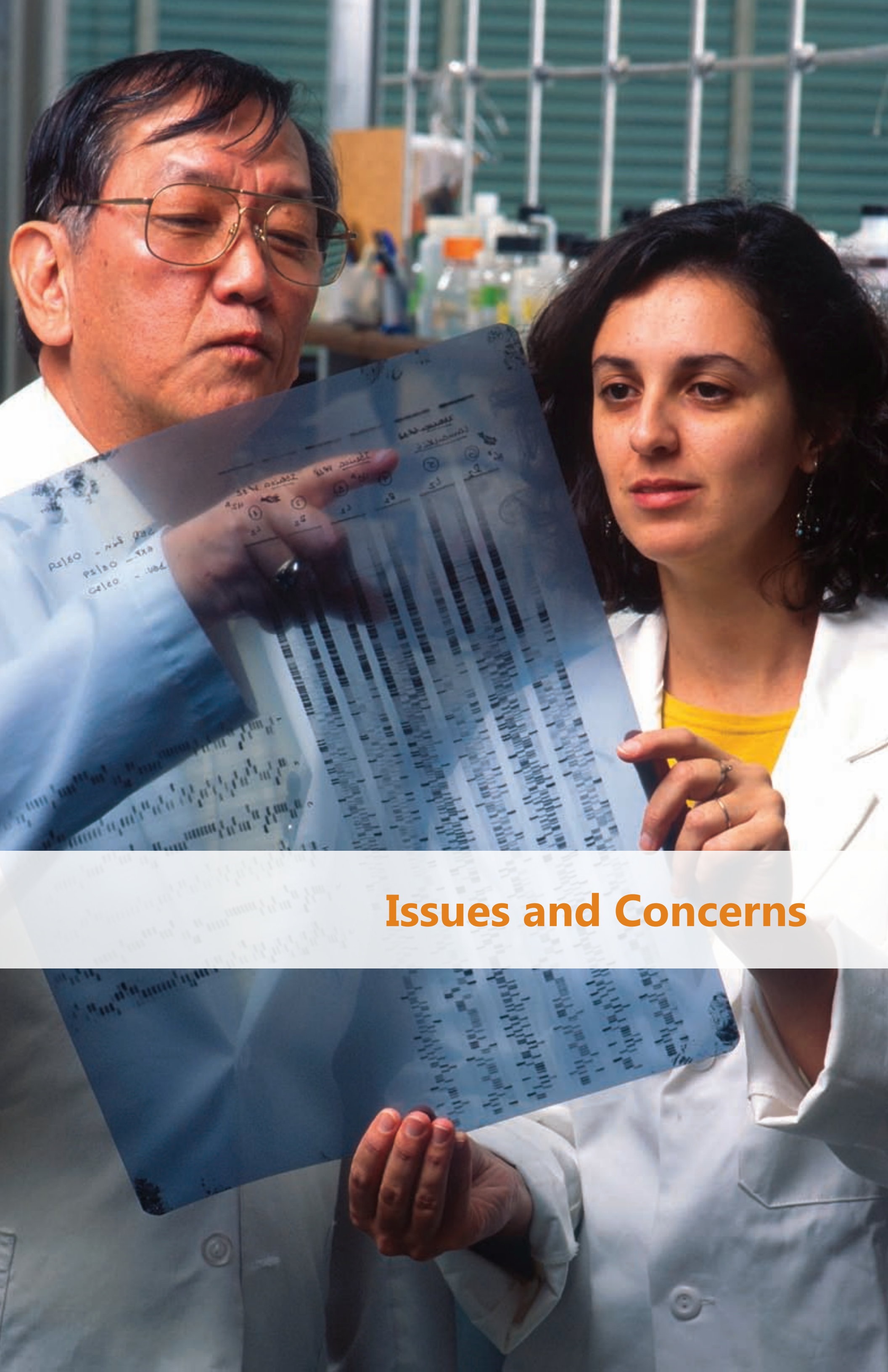

Issues and Concerns

Cartagena Protocol on Biosafety

While advances in biotechnology have great potential to improve human well-being, the technology must be developed with adequate safety measures. The Cartagena Protocol on Biosafety is a legally binding global protocol to the Convention on Biological Diversity (CBD). It seeks to contribute to ensuring the safe transfer, handling and use of living modified organisms (LMOs) created through modern biotechnology.

The Protocol was named in honor of Cartagena, Colombia, where negotiations were expected to conclude in February 1999. One year later, on January 29, 2000, the Protocol was finalized and adopted in Montreal, Canada by unanimous consent with 135 countries present. It was entered into force on September 11, 2003.

What is the Protocol's objective?

Article 1 of the Protocol states that it aims to "contribute to ensuring an adequate level of protection in the field of the safe transfer, handling and use of living modified organisms resulting from modern biotechnology that may have adverse effects on the conservation and sustainable use of biological diversity, taking also into account risks to human health, and specifically focusing on transboundary movements." In short, it seeks to protect biodiversity from the potential risks of living modified organisms (LMOs) resulting from modern biotechnology.

What does the Protocol cover?

The Protocol covers the "transboundary movement, transit, handling and use of all living modified organisms that may have adverse effects on the conservation and sustainable use of biological diversity, taking into account risks to human health."

It does not cover:

- Products derived from LMOs (e.g. paper from GM trees)
- LMOs, which are pharmaceuticals for humans that are addressed by other relevant international agreements or organizations

What does the Biosafety Protocol do?

- It assists developing countries in building their capacity for managing modern biotechnology.
- It creates an Advanced Informed Agreement (AIA) procedure that requires exporters to seek consent from importing countries before the first shipment of LMOs meant to be introduced into the environment (e.g. seeds for planting, fish for release, and microorganisms for bioremediation).
- It establishes an internet-based “Biosafety Clearing-House” to help countries exchange scientific, technical, environmental and legal information about LMOs.
- It requires bulk shipments of LMO commodities, such as corn or soybeans that are intended to be used as food, feed or for processing, to be accompanied by documentation stating that such shipments “may contain” LMOs and are “not intended for intentional introduction into the environment”.
- The Protocol includes a clause that makes clear the Parties’ intent that the agreement does not alter the rights and obligations of governments under the World Trade Organization (WTO) or other existing international agreements.

What does the Biosafety Protocol not do?

- The Protocol does not address food safety issues. This is addressed by experts in other international fora.
- The Protocol does not require segregation of bulk shipments of commodities that may contain living modified organisms.
- It does not require consumer product labeling.
- It does not subject shipments of bulk commodities to the Protocol’s AIA procedure.

As of March 2015, 170 countries have ratified the Protocol. When a country signs the Protocol, it signifies its general support for the principles in the Protocol and commits to take the steps necessary to consider and pursue its ratification. The Protocol only becomes legally binding when a country deposits an instrument of ratification with the United Nations.

Key Features of the Protocol

Advanced Informed Agreement (AIA)

The Protocol's main mechanism is its Advanced Informed Agreement (AIA) requirement. It is a procedure that must be followed before the first intentional transboundary movement of an LMO into the environment of the importing country. The exporter must provide a notification to the importing country containing detailed information about the LMO, previous risk assessments of the LMO and its regulatory status in the exporting country. The importing country must acknowledge receiving the information within 90 days and whether the notifier should proceed under a domestic regulatory system or under the Protocol procedure. In either case, the importing country must decide whether to allow the import, with or without conditions or deny it within 270 days.

What is not subject to the AIA requirement?

- Consecutive shipments. The Protocol's AIA only covers first time shipments.
- LMOs not intended for release into the environment such as commodities, LMOs in transit, and LMOs destined for contained use.

Biosafety Clearing-House (BCH)

The BCH is a website administered by the Secretariat to the Convention (http://bch.biodiv.org). It was established to:

- facilitate the exchange of scientific, technical, environmental and legal information on, and experience with LMOs; and
- assist Parties to implement the Protocol.

Examples of information contained in the BCH include: any existing laws, regulations, or guidelines for implementation of the Protocol, summaries of risk assessments or environmental reviews of LMOs, and final decisions regarding the importation or release of LMOs.

Risk Assessment

The Protocol requires that decisions on proposed imports be based on risk assessments.

Risk assessments must be undertaken in a scientific manner based on recognized risk assessment techniques, taking into account advice and guidelines developed by relevant international organizations.

Lack of scientific knowledge or scientific consensus must not necessarily be interpreted as indicating a particular level or risk, an absence or risk, or an acceptable risk.

Risks associated with LMOs or products thereof should be considered in the context of risks posed by the non-modified recipients or parental organisms in the likely potential receiving environment.

Risk assessment should be carried out on a case by case basis.

Capacity Building
The Protocol promotes international cooperation to help developing countries acquire resources and capacity to use biotechnology safely and regulate it efficiently. It does this by encouraging member governments to assist with scientific and technical training to promote the transfer of technology, knowledge, and financial resources. Governments are also expected to facilitate greater involvement of the private sector.

Public Awareness
Member governments must commit themselves to promoting public awareness, insuring public access to information, and public consultation. The Protocol recognizes that national measures are important to make its procedures effective. Nations must also take measures to prevent illegal shipments or accidental releases of LMOs.

Useful websites

CBD. 2015. Biosafety Clearing-House. https://bch.cbd.int/.
CBD. 2015. The Cartagena Protocol on Biosafety. https://bch.cbd.int/protocol.

Intellectual Property Rights and Agricultural Biotechnology

One of the main features of modern agricultural biotechnology (agri-biotech) is its increasing proprietary nature. Unlike the agricultural sciences of the past, which came out of publicly funded labs, new biotechnologies are protected by patents and other intellectual property rights (IPRs). The ownership of IPRs in agri-biotech is now an issue in the development of products and the transfer of the technology to developing countries. Scientists now need to consider IPRs as an important factor in their research, especially where the aim is product development.

What is Intellectual Property?

Intellectual property represents products of the mind or intellect. They are ideas that, when converted to tangible forms, can be protected. Examples of intellectual properties include inventions, computer software, publications, videotapes, music, and plant varieties.

Developing such products usually requires a great deal of time and financial investment. Therefore, the inventor usually seeks a return on his effort by acquiring IPRs. They allow the inventor to restrict the use of the intellectual property, i.e., no one is allowed to use, manufacture, grow, sell or offer to sell the invention without permission. Several forms of this protection exist and they include copyright, trade secret, trademarks, plant breeder's rights, and patents. Intellectual property is a tool to foster innovation.

What are the functions of IPR?

IPRs are intended to promote research and development by providing incentives for investment in the creative process and encourage access to inventions produced elsewhere.

IPRs and developing countries

Patents, plant breeder's rights and trademarks are awarded by national governments, and the protection is valid only in countries in which they are issued. Thus, to obtain protection in several countries, rights must be applied for and awarded in each. On the other hand, copyright and trade secrets are not country specific.

At present, many key technologies used in the development of agri-biotech products appear to be unprotected in developing countries. Furthermore, anyone is free to use technologies in crops that are developed, produced, and consumed in countries where the technology is not subject to local IP protection. IP problems, however, may arise when these crops are subsequently exported to countries in which the technologies are protected by IPRs. The development time should also be taken into consideration since patents might be issued in the country by the time the product is developed. It is therefore necessary for scientists in developing countries to be aware of the IP issues and develop strategic plans in handling these IP concerns.

Promoting transfer of agri-biotech to developing countries

Crops grown for subsistence use in developing countries and the technologies that are used to develop such crops are clearly of little commercial interest to the private sector. Thus, donating proprietary technologies to develop such crops is a realistic possibility, and in fact is already happening. However, developing country scientists must remember that technology transfer involves a lot more than simply signing a license or a material transfer agreement for a product. Both technology donor and recipient must be aware of the IPR issues involved in the technology and there will often be a need for partnerships in which there is mutual trust among all parties (Kratigger, 2002).

Developing countries frequently lack the required IP management capacity and resources to perform product clearance analyses and evaluations that facilitate the legitimate import, use and/or export of technologically advanced products (Kowalski, et al., 2002). Thus, to help transfer of appropriate agri-biotech to developing countries, capacity building in IPR management is of vital importance from both the donor and the recipient side. This can involve educating research staff and research administrators on the basic principles of IPR management; and using different patent databases as well as scientific databases as information sources.

How do you protect your rights?

The main ways to protect intellectual property rights are patents, copyrights, trademarks, geographical indications, and trade secrets. In many countries, plant variety protection or plant breeders' rights are offered (Krattiger, et al., 2007). Most relevant forms of IP protection in plant breeding are plant breeder's rights and patents.

Plant Breeder's Rights

Plant breeder's rights (PBRs) are used to protect new varieties of plants by giving exclusive commercial rights for about 20-25 years to market a new variety or its reproductive material. The variety must be novel, distinct, uniform, and stable. This protection prevents anyone from growing or selling the variety without the owner's permission. Exceptions may be made, however, for both research and use of seed saved by a farmer for replanting.

Patent

A patent is an exclusive right given to an inventor to exclude all others from making, using, selling or offering to sell the invention in the country that granted the patent right, and importing it into that country. In agricultural biotechnology, patents may cover, for example, plant transformation methods, vectors, genes, etc. and in countries that allow patenting of higher life forms, transgenic plants or animals.

Patents are the most critical form of protection for agricultural biotechnology and considered to be the most powerful in the IP system. Patents are temporary, generally about 20 years, and are country specific (Binenbaum et al., 2000).

Conclusion

Publicly funded research institutions should build up their capacity to manage intellectual properties that they procure and those that they generate. Knowledge of IPRs will help developing country scientists determine if information about a particular technology is already part of the public domain and therefore freely available. Moreover, IPs generated by the public sector can be considered assets that can be exchanged for private sector-owned IPs or used as bargaining chips in technology transfer negotiations. Partnership between the private and public sectors in technology development through sharing of knowhow and IP can hasten technology transfer and acquisition on both sides.

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Labeling GM Foods

The debate over foods derived from genetically modified (GM) crops often touches on the subject of labeling. Many consumers argue and insist on their right to know what they are eating and their right to choose. We hear it all the time: "Why not label these foods if you are so sure of their safety?" or that "Consumers should have a choice about what they are eating." As a result, many governments have begun to heed these suggestions and have either implemented labeling regulations or are working on them.

Unfortunately, while the questions seem simple, the issue is not, especially if the starting point of labeling includes the process rather than the final product. Issues such as safety, cost, truth in advertising, choice, fairness, science, trade-barriers, regulatory responsibility, accountability, legal liability, among others are involved.

Requirements for implementing labeling policies

Standards, testing, certification, and enforcement

Before any labeling rules can be implemented, governments would have to set up standards and services to conduct testing of the presence of GM ingredients; certification; and ensure that the quality standards are clear and achievable.

While it is easy to detect GM ingredients in products where the GM product is the main ingredient (like tofu or popcorn), it would not be so easy to detect them in processed products like oils, sugars and starches, which no longer contain any novel DNA or proteins.

On another note, much of the food that is bought and consumed in developing countries is not packaged and consequently not labeled. Examples are soybean milk from a street vendor or fresh fruits and vegetables from the market.

Another issue that regulators have to grapple with is the wording: ideally a label should not prejudice the consumer for or against the product.

There is also the issue of whether the label would be useful or educational. To a homemaker who has heard little about the debate on GM food, a label that reads, “Made from genetically modified soybean” or “Grown from seed obtained through modern plant biotechnology” may create more confusion.

Current labeling regulations

Current regulations are based on the chemical characteristics of the food product and not on the way the product was made. For example, labeling regulations only require labels for foods (whether GM or not) if there is a change in nutritional composition or if an added component is toxic or allergenic.

More than 40 countries have adopted labeling regulations for GM food. However, labeling policies differ widely in their nature, scope, coverage, and degree of enforcement. Only a few developing countries have introduced labeling laws and ever fewer have implemented them.

The common feature among countries with labeling laws is the requirement to label products derived from GM crops that are not substantially equivalent to their conventional counterparts. An example would be nutritionally enhanced GM crops. For products that are substantially equivalent to conventional products such as from 1st generation GM crops, there is a large international heterogeneity in labeling regulations.

Countries with voluntary labeling (e.g., Canada or Hong Kong) dictate rules that define which foods are called GM or non-GM. Food companies are allowed to decide if they want to use such labels on their products. Australia, the European Union, Japan and China follow mandatory labeling. Food handlers from food processors retailers and sometimes food producers display whether the targeted product/ingredient contains or is derived from GM materials. Regulations though differ widely based on coverage, threshold level for labeling of GM ingredients, and labeling content.

Implications of labeling food

How will it affect world and regional trade?

As the production and trade of GM crops increase, labeling programs will allow countries to tailor policies to their own needs. For example, a country can take its time to allow GM crops to be grown within its boundaries, but allow the import of such crops and food products as long as they are labeled. Several key trading partners of the U.S. have recently instituted mandatory labeling policies and as a result, will only allow imports of GM products from the U.S. if they are labeled. This is most likely to create political tension with the U.S. and other similar countries that are exporting GM food products. Finally, the GM labeling issue will also be looked at as a possible trade barrier.

What is the cost of labeling?

It is not simply the cost of ink and stamps. Auditing must be done from the very beginning of the food production stream, starting with the seed companies, and following through to the farmers, the grain companies, the food processors, the distributors, and marketers. The huge cost is associated not with putting a label on but with keeping it off. The non-GM food producer must document every step of the process, going back not to the farmer, but to the seed supplier. Verification assays to test positive cost less than assays to test negative because the positive needs only one positive score on one assay to complete the verification but a non-GM label requires a series of negatives on every assay.

Studies on the cost of mandatory labeling in Canada, Australia, the United Kingdom, the Phillippines, and in the U.S. have shown that main costs estimates range from \$0.20 up to \$10 or even \$20 per capita per year. In a developing country such as the Philippines, mandatory labeling would result in a 11-12% production cost increase or an additional 10% increase in consumer prices.

Therefore, any form of labeling, whether for GM or non-GM products, will entail additional cost. This will initially be borne by the producers but would probably be passed on to the consumers. Will consumers be willing to pay higher prices?

Conclusion

The issue of labeling GM foods is a complex issue that has yet to be resolved. What is clear, however, is that some kind of labeling policy will be adopted by most countries. Right now, the decision to label GM products is not so much related to the actual safety of the product, but rather to the "fear" alluded to such products. The presence of a GM label should not imply that the product is less safe or is significantly different since all GM foods have to meet safety standards before being approved for sale.

The only way to develop and maintain a labeling system that is truthful, not misleading, and verifiable is to ensure it is based on objective criteria, such as the actual composition of the food, and not on the method of manufacture.

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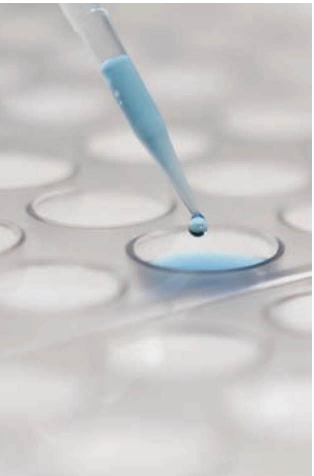
Ethics and Agricultural Biotechnology



Similar to other emerging technologies, biotechnology has instigated worldwide debate and confusion as a result of mixed messages from various people — be they scientists, academics, activists, industry, religious representatives or consumer bodies. It has stirred conflicting ideas and opinions and has polarized sectors not only among stakeholders but even between countries.

While agriculture has long been a topic of philosophical, religious and political reflection, it is only in the late 20th century that systematic thinking about the values and norms associated with the food system, such as farming, food processing, distribution, trade, and consumption, began to be discussed in the context of agricultural ethics.[1]

In 2000, the Council of Europe Parliamentary Assembly recommended that it was increasingly important to include ethical considerations centered on humankind, society and the environment in deliberations regarding developments and applications in biotechnology, life sciences, and technology. A year later, the United Kingdom's Royal Society Report asserted that "public debate about genetically modified food must take account of wider issues than the science alone."[2]

What is agricultural ethics?

In general, 'ethics' is defined as the ideals, values or standards that society uses to judge whether an issue or thing is acceptable and justifiable and determines responsibility and justice.[3]

Ethics in agricultural biotechnology encompass value judgments that cover the production, processing, and distribution of food and agricultural products. The Food and Agriculture Organization of the United Nations (FAO) asserts that ethical values determine its reason for being — these being the values for food, enhanced well-being, human health, natural resources, and nature.[4]

What are some ethical issues raised about agricultural biotechnology?

Many of the ethical issues that form part of the biotechnology debate can apply also to food and agricultural systems in general. Accepting the need to understand and tolerate societal norms or beliefs, many statements of concern are often general and broad with little explanation about what makes them disagreeable or wrong. The following are examples of issues.[1,2,3,4]

"Playing God"

Genetic modification is said to involve human intervention into creation and hence, is an unnatural act. Often viewed as a religious question, it avers that the technology is "so intrusive to life processes that they amount to a form of disrespect for humanity's proper relationship to nature, a form of playing God." Some religions ascribe a particular "essence" to each living organism and hence, connect the concept of gene with the idea of essence. Others believe that biotechnology disrupts natural order and violates the limits of what humans are ethically permitted to do. Alternatively, there is the view that science and progress are good things and are God-given faculties to help mankind support life and better manage the environment.

Islamic scholars note that Islam and science are complementary and Islam supports beneficial scientific innovations to address food security.[5] The Pontifical Council for Justice and Peace released the Compendium of the Social Doctrine of the Church in 2004 which is an "overview of the fundamental framework of the doctrinal corpus of Catholic social teaching." Biotechnology is mentioned as having powerful social, economic, and political impact but that it should be used with prudence, objectivity, and responsibility.[6]

General welfare and sustainability

A central issue is whether the technology considers the pursuit of the greatest good together with the concept of sustainability for farmers and the environment. While a technology can provide more food it should not be to the detriment of the environment or to human health or disrupt traditional behavioral systems. In like manner, it is an ethical issue if food that can provide more and better nutrition is not made available to those who need it most. Hence, not to use a technology that has potential to improve the quality of lives of people is also a moral issue. As an environmental issue, questions raised have to do with concerns regarding environmental protection, sustainable use of biodiversity, economic growth and social equity.

Distribution of benefits and burdens

A concern particularly in developing countries is the concept of just distribution. Questions have to do with whether the products produced by the technology will be able to provide for those who really need them and whether they will generate wealth for the society as a whole. A technology's ability to increase or decrease the gap between the rich and poor renders it an ethical issue. This includes allegations that products derived from modern biotechnology are being introduced by private companies that have an obligation to make profits. Also up for discussion is whether a technology, while able to increase technical employment might eliminate subsistence labor as a result of replacing cultural operations.

Other concerns include exploitation or control over genetic resources, consumers' choice and rights, and use of genetically modified animals.

How do we deal with ethical issues?

FAO recognizes that there is no single set of ethical principles sufficient for building a more equitable and ethical food and agricultural system.[4] However, it recommends several actions that individuals, states, corporations and voluntary organizations in the international community can take. These include among others: creating the mechanisms to balance interests and resolve conflicts; supporting and encouraging broad stakeholder participation in policies, programs, and projects; encouraging individuals, communities and nations to engage in dialogue, and ultimately, to do what is ethical; and developing and disseminating widely the information and analyses necessary to make wise and ethical decisions.

Conclusion

Despite the diversity of ethical issues in agricultural biotechnology, there is a need to understand beliefs and doctrines as this allows coexistence within and across societies, and prevents social conflict. A technology's acceptance is based not only on technological soundness but on how it is perceived to be socially, politically, and economically feasible from the viewpoint of disparate groups. An understanding of ethics helps determine what information is needed by society and how to deal with different opinions. A process of negotiation based on trust is essential to enable stakeholders to participate in debates and decision making.

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Kenya National Biotechnology Development Policy Highlights

The government of Kenya has developed a comprehensive national policy to guide research, development and commercialization of modern biotechnology products. The policy, which was approved in September 2006, has been the result of several years of work involving all major biotechnology stakeholders nationally, internationally, and relevant government departments.

The policy charts the vision of the Kenyan government towards the development and safe application of biotechnology. It provides those developing and applying the technology with a clear framework under which to operate. The policy commits the government to give priority to the provision of relevant institutional, infrastructural and legislative framework and, in particular, the enactment of new legislation on biosafety.

Objectives of the policy

- Prioritize, promote, and coordinate research in basic and applied bio-sciences
- Promote sustainable industrial development for production of biotechnology derived products
- Create enabling administrative and legal frameworks for biotechnology development and commercialization
- Develop mechanisms for the provision of sustainable funding for biotechnology research and products development
- Support and facilitate capacity building on all aspects of biotechnology including intellectual property access and protection, biosafety and bioethics
- Support the development and retention of human resources in science, innovation and biotechnology
- Stimulate collaboration among public, private sectors and international agencies in order to advance biotechnology both locally and internationally
- Promote public understanding of the potential benefits and address stakeholder concerns/issues on modern biotechnology

Scope of the policy

The policy outlines six priority areas of focus:

1. **Agricultural Biotechnology**
   Biotechnologies to develop new plant varieties with beneficial genetic traits for pest and disease resistance, improved nutritional value, tolerance to drought and salinity. Special attention will be paid to conservation of germplasm of traditional and wild crop plants.
   - Animal reproductive biotechnologies such as artificial insemination, embryo transfer, genetic improvement of local breeds, and somatic

cell nuclear transfer (cloning) techniques. Special attention will be paid to the development of livestock that are resistant to diseases, have improved meat, milk or wool quality, can increase proteins in their milk or meat (biopharm animals), or which have characteristics that are environmentally friendly

- New plant and animal diagnostic products, improved animal vaccines, biological pesticides, herbicides and fertilizers

2. Education

Reviewing of curricula at all levels to promote the spirit of scientific inquiry by encouraging independent student projects, exposing students and teachers to biotech activities in Kenya and internationally through study tours, expert guest lectures; and promoting acquisition of entrepreneurial skills.

- Strengthening the teaching of biosciences at the formal education level
- Attracting and retaining talent in biosciences
- Developing scientific and related infrastructures
- Spearheading formal and informal public education and awareness creation programs

3. Bioresources

Creation of research fund to facilitate molecular characterization and bioprospecting for novel products for development and industrial production.

- Establishment of national culture collection centers for the preservation and utilization of economically beneficial microorganisms.
- Accelerate the establishment of viable *in situ* and *ex situ* (Gene banks) conservation centers.
- Focused exploitation of fauna, flora and microbes in marine and extreme habitats for novel genes for development of osmo tolerant crops, enzymes, biopolymers, marine pollution biosensors, bioactive molecules, etc.

4. Environmental Biotechnology

Monitoring of environmental pollution

- Eco restoration of degraded habitats
- Afforestation and reforestation
- Bioremediation of wastes
- Control of biological invasions
- The potential for value-added products from biomass

Applications requiring use of modern biotechnology for all purposes, will be subject to approval by the designated authority,

5. Medical Biotechnology

Basic and applied research in bioinformatics, molecular and cellular biology, genomics, proteomics, stem biology (strictly using ethically obtained stem cells only), and other new platform biotechnologies as appropriate

- Development of molecular diagnostics, recombinant vaccines, and drug delivery systems

- Development of traditional herbal medicines into superior industrial therapeutic products
- Screening of biodiversity components for bioactive compounds for value added therapeutic products
- The policy outlaws any activities or research dealings involving human cloning, use of unethically procured stem cells, and the introduction, use or release of the "Terminator Technology" and associated products into Kenya

6. Industry and Trade

Develop initiatives that will attract major investment in biotechnology research and product development from local and international companies or institutions.

- Promote industrial skills development.
- Provide a conducive environment for small and medium size biotechnology products businesses.
- Ensure high quality standards, competitiveness of products on local and international markets.

Key policy and recommendations

I. Prioritization and Coordination of Research and Development

The policy recommends establishment of a National Biotechnology Enterprises Programme that will consist of a National Commission on Biotechnology, a National Biotechnology Education Centre and a National Biosafety Authority.

Functions of the National Commission on Biotechnology will be to consolidate and maximize on available resources of institutions engaged in training and R&D through:

- Identification and implementation of national priority areas for R&D
- Provision of advice/guidance on and/or supervision of the allocation of primary resources and responsibilities to public R&D institutes and universities
- Tracking and evaluation of inventions, patents, and commercialization of discoveries
- Identifying and linking R&D centers of excellence and the private sector.

The National Biotechnology Education Centre will:

- Coordinate and facilitate training and knowledge-sharing
- Develop and maintain bioscience research, innovation and biotechnology database
- Develop and maintain a National culture collection

A National Biosafety Authority will be responsible for safe acquisition, development and commercialization of biotechnology and its products thereof. The authority will be the central coordinating and implementation body and will work together with other government regulatory bodies to ensure adherence to laws and regulations.

II. Public Education and Awareness Creation

- Creation of public awareness on biotechnology issues and investment opportunities
- Access to information held by public authorities
- Public participation in decision making process
- Access to judicial and administrative provisions

III. Public Protection and Support

- Protecting Intellectual Property Rights (IPR) is a critical aspect of biotechnology innovation, and ensuring effective public and private sector participation in research and product development.
- The Government recognizes the existing policies and legislation on protection of traditional knowledge and resources.

IV. Infrastructure, Facilities and Equipment

- The National Biotechnology Enterprises Programme to put in place mechanisms to create linkages and networks among public research institutes and universities for optimum access and utilization of available resources
- Enhancement of public/private partnerships
- Support initiatives for the establishment of biotechnology parks at R & D institutions as incubators to stimulate the growth of small and medium size businesses with potential to mature into high technology companies

V. Financial and Business Support

Create incentives to encourage partnerships between public research institutes and universities, and the private sector for the purpose of attracting private sector investment in biotechnology based start up firms. Incentives include but not limited to subsidies on private sector capital investment and tax exemptions.

- Waiver of taxes on research materials and equipment
- Encourage specialized technological financing agencies to provide loans to firms or consortia and research institutions
- Direct public budgetary allocation to biotechnology research and development

For more information: Biosafety Office www.biosafetykenya.co.ke.

Communicating Crop Biotechnology

Crop biotechnology, while merely one of the many possible scientific options to improve agricultural productivity, has triggered increased interest in its consistent and substantial benefits. At the same time, it has sparked debate on its perceived risks and safety and is often caught in a maelstrom of controversy. Diverse issues like scientific, political, economic, ethical, cultural, and even religious viewpoints are being espoused by different stakeholders. Crucial therefore to balancing issues and concerns surrounding biotechnology is adequate science-based, authoritative information to enable various stakeholders to engage in an objective and transparent debate. Mutual understanding and dialogue will enable the global community to understand the attributes of crop biotechnology and assure acceptable by the public.

Traynor, et al. identify some specific objectives for public communication: make evident to decision makers that modern biotechnology can be an effective tool for increasing agricultural productivity, and thereby economic growth, without imposing unacceptable risk to the environment or human and animal health; and enable members of the public to make informed decisions about appropriate uses of biotechnology by providing accurate information about benefits, risks, and impacts. [1]

Why is communication important?

Communication is one of several key variables needed to create an enabling environment for biotechnology. Efforts to encourage stakeholders to participate in evidence-based discussions are needed. These will allow decisions to be made and to build consensus regarding the acceptance and adoption of technology. The public involvement process is then able to introduce issues beyond the boundaries of science such as socio-cultural, political, and ethical concerns.[2, 3]

Saner enumerates reasons why we need to involve the public: potentially improve public policy, a more informed and engaged public, more solid support for regulatory decisions, and greater public confidence in government.[4] Communication therefore include these activities: inform or educate to help understand a policy or program; gather information to anticipate communication challenges; facilitate discussion among stakeholders; engage citizens for shared agenda setting and generate options; and partnering or reaching agreement among stakeholders.

What are the steps in implementing communication activities?

There are five important steps in implementing communicating activities.[5] The process is cyclical, as it involves a continuous flow of reassessment and refinement. Versoza enumerates these steps as:

- ***Assessment.*** This stage involves obtaining information to guide the communication strategy. It identifies the behaviors desired, key messages, audiences or stakeholders to reach, the communication channels to reach the audience, and specific units to implement the communication activities.
- ***Planning.*** A clear course of action is determined on the basis of the assessment earlier done. Decisions are made with regards desired behaviors, key messages, audiences, communication channels, and activities including supporting elements such as budget, timeline, communication research plan, and a capacity building component.
- ***Material development and pretesting.*** Production of communication materials entails working with the audience to develop messages that will be effective with them. Hence, messages must be clear and easy to understand, and culturally sensitive.
- ***Implementation.*** The delivery and distribution of communication materials whether through print, radio or television, or through interpersonal communication means depends not only on quality and timeliness, but also on availability of good supporting services.
- ***Monitoring and evaluation.*** These are carried out simultaneously with implementation to determine audience response to messages, and subsequent changes in knowledge, attitudes, beliefs and practices. This process enables mid-course corrections and identifies new opportunities to improve the communication component. The final evaluation enables learnings to be used for future communication programs.

What communication activities can be implemented to increase greater awareness and understanding of biotechnology?

Biotech communication strategies must be linked with each country's cultural and political climate. It is driven by a number of interrelated factors: knowledge level, awareness of benefits, confidence, and trust.

A strategic and complementary combination of interpersonal communication and different mass media modalities is recommended. Interpersonal communication is needed to achieve acceptance and use of technology while mass media help promote awareness, knowledge, and understanding. Personal interfaces allow people to interact in close proximity, use sensory channels to relay messages, and receive immediate feedback. Use face-to-face communication with multi-media strategies (publications, electronic-based formats, videos, and exhibits). The possibilities and combinations are endless and are limited only by communicators' imagination and willingness to think out-of-the-box.

What are some insights in communicating biotechnology?

Experiences learned from communicating biotechnology through the years have given rise to several lessons.[6,7] These include:

- Communication is not merely a one-way process of dishing out information to people based on the assumption that lack of understanding stems from inadequate information or that ample information can compel action. Rather, it involves social negotiation and dialogue between and among varied audiences.
- In embarking on any science communication initiative, it is important to take stock of the current environment for biotech taking into consideration scientific developments, political support, role of key players vis a vis biotech, and influence of stakeholders in decision-making process. There is a need to identify stakeholder issues, key information sources and gaps, and barriers and opportunities to biotechnology acceptance in the country.
- Organizations involved in communicating biotechnology should not be merely information centers. They should strive to be significant players in the development of enabling environments for informed decisions.
- A strong and effective cadre of science communicators is essential particularly among those who see the need for transparent and

science-based discussion and debate. Capacity building in science communication, media relations, public engagement, science popularization, and media development and production is crucial.
- There is a need to identify and nurture champions from different stakeholder groups (policy makers, scientists, academics, regulators, farmers, and the media). These champions should be well-informed, have high credibility, and are willing to share the attributes of the technology among their peers.
- Public attitude towards technology is often based on values more than information itself. These values include high trust in science and the regulatory system, credibility, and freedom of choice. Thus, it is more effective to frame communication around a value(s) rather than on the technology.

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