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# Biotechnology & Genetically Modified Organisms

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## ABSTRACT

One of the many applications of modern biotechnology is the use of GMOs in the food production chain. GMO (Genetically Modified Organisms) means remove one or more genes from the DNA of another organism, such as a bacterium, virus, animal, or plant and "recombine" them into the DNA of the plant they want to alter. Therefore do not fall in the category GMO living beings who have undergone genetic changes through spontaneous processes and through intersections, but only those for which it was made use of genetic engineering, more specifically to the hybridization techniques and selection, mutagenesis and selection. Genetic modification is based on a theory called the Central Dogma, which asserts that one gene will express one protein. However, scientists working with the United States National Human Genome Research Institute discovered that this wasn't true, that genes operate in a complex network in ways that are not fully understood. This finding undermines the entire basis for genetic engineering. Today, GM foods are as controversial as ever, particularly given that a large amount of research studies are conflicting and many have been presented in slanted or exaggerated ways. It is difficult for the consumer to sort through the barrage of conflicting information. When a person does not fully understand a concept and has mixed information, the tendency can also be to want to avoid it entirely. For now though, the best approach is one of education and information, which will allow you to contribute to a safer and more positive future of GM foods.

**Keyword:** GMO, DNA, PCR, ELECTROPORATION.

## INTRODUCTION

Biotechnology encompasses a wide range of technologies and they can be applied for a range of different purposes, such as the genetic improvement of plant varieties and animal populations to increase their yields or efficiency; genetic characterization and conservation of genetic resources; plant or animal disease diagnosis; vaccine development; and improvement of feeds. Some of the technologies may be applied to all the food and agriculture sectors, such as the use of molecular DNA markers or genetic modification, while others are more sector-specific, such as tissue culture (in crops and forest trees), embryo transfer (livestock) or triploidization and sex-reversal (fish). Genetically modified crops (GMCs, GM crops, or biotech crops) are plants used in agriculture, the DNA of which has been modified using genetic engineering techniques. In most cases, the aim is to introduce a new trait to the plant which does not occur naturally in the species.

Examples in food crops include resistance to certain pests, diseases, or environmental conditions, reduction of spoilage, or resistance to chemical treatments (e.g. resistance to a herbicide), or improving the nutrient profile of the crop. The first modern GMO was obtained in 1972 by Stanley Cohen (Stanford University School of Medicine) and

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Herbert Boyer (University of California, San Francisco). The two researchers, thanks to the combined use of new molecular biology techniques that were developing in different laboratories, managed to clone a frog gene into the bacterium E. Coli, demonstrating that it was possible to transfer genetic material from one organism to another fact of breaking down the species-specific barriers. In 1974, the scientific community autoimpose an international moratorium on the use of recombinant DNA technology, to allow time to assess the status of the new technology and the possible risks through a precautionary approach. The Asilomar Conference held in 1975, addressed the problem of the safety of experiments with recombinant DNA technology (genetic engineering) which began in those years The Asilomar Conference in 1975, promoted by the scientists themselves, was an example of self-regulation by the scientific community held in California. The Conference concluded that the experiments on recombinant DNA could proceed as long as under strict guidelines, then edited by the National Institute of Health, and accepted by the scientific community. Our guidelines, published for the first time in 1976 and later updated, still are the foundation that inspires all laboratory research involving gene transformation experiments. Examples in non-food crops include production of pharmaceutical agents, biofuels, and other industrially useful goods, as well as for bioremediation [1]. The first genetically modified crop plant was produced in 1982, an antibiotic-resistant tobacco plant [2]. The first field trials occurred in France and the USA in 1986, when tobacco plants were engineered for herbicide resistance [3]. In 1987, Plant Genetic Systems (Ghent, Belgium), founded by Marc Van Montagu and Jeff Schell, was the first company to genetically engineer insect-resistant (tobacco) plants by incorporating genes that produced insecticidal proteins from Bacillus thuringiensis (Bt) [4]. Monsanto was one of four groups to introduce genes into plants (1983), [5] and was among the first to conduct field trials of genetically modified crops, (1987). It was one of the top 10 U.S. chemical companies until it divested most of its chemical businesses between 1997 and 2002, through a process of mergers and spin-offs that focused the company on biotechnology. Monsanto become one of the major industrial empires on the planet because he carried out anything less than a large-scale production some of the most dangerous products of modern times. One of these was the PCBs (polychlorinated biphenyl), which have been used as coolants and lubricants, especially damaging and devastating to human health. Recent studies indicate that maternal consumption of PCB-contaminated fish can cause disturbances in reproductive parameters and neurobehavioral and developmental deficits in newborns and older children. For, in the course of many years of marketing its products—PCBs, herbicides, dioxin, bovine growth hormones, Roundup—Monsanto was fully aware of their harmfulness. Thanks to extensive research by Marie-Monique Robin, 'found that the company knew the toxicity of this product, but behind this was engine of business. In Monsanto's position outside democratic control, it is hard to tell whether it is commercial blindness, scientific arrogance, or pure and simple cynicism that dominates. In 2003 in Taiwan was sold the first GMO animal in the household: a hundred aquarium fish made fluorescent by inserting jellyfish genes.

In December 2003 the sale of fluorescent fish has also been allowed in the United States, after the Food and Drug Administration declared no relevance for food of these fish while it is still prohibited their sale in Europe. The issue of GMOs is still hotly debated today, not only at national level but also at international and EU level, partly because of the fragmentation and diversity of the involved decision-making centers, the difficult balance between the requirements of protection of collective security and the guarantee of fundamental rights and freedoms guaranteed by the Constitution and the incompleteness of scientific knowledge.

## METHOD

Genetically Modified Organisms the term is often used in a reductive by the media to refer only genetically modified plants. In fact, the great family of GMOs is more nourished. But why should we genetically modify a plant? What would be the consequences? And how could we do it? Transgenic (GM) plants are those that have been genetically modified using recombinant DNA technology. DNA, or deoxyribonucleic acid, is the hereditary material in humans and almost all other organisms. Nearly every cell in a person's body has the same DNA. DNA bases pair up with each other, A with T and C with G, to form units called base pairs. Each base is also attached to a sugar molecule and a phosphate molecule (**Figure 1**).

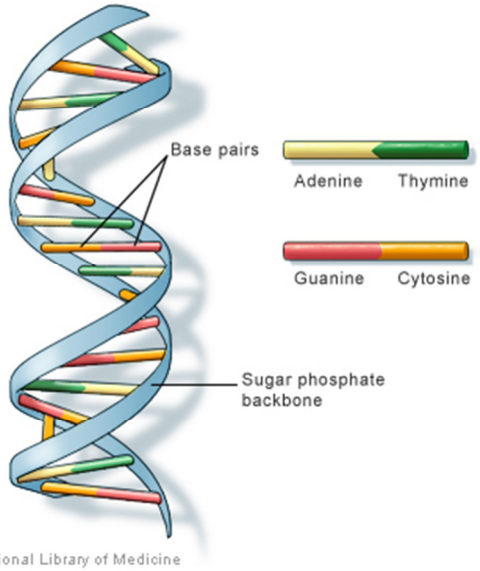


**Figure 1**. Credit: U.S. National Library of Medicine

The DNA of an organism can then be modified by inserting sentences (genes) of other species, to allow the cells to produce a protein of interest. This protein will confer to the body - that we will call now genetically modified - a new feature: the resistance to pests, to the production of vaccines and so on. The main aims are to obtain specimens "best". In the case of plants for example a transgenic [6] product can:

- be stronger and avoid the use of pesticides
- be less susceptible to pollutants (such as carcinogenic aflatoxin to humans)
- to obtain a better productivity index
- have better nutritional characteristics (e.g. greater wealth of vitamins or lower content of uninteresting or even harmful substances).

The procedure used to produce plant GMO can be simplified into four steps **Figure 2:**

- *Isolate* the genetic trait.
- *Insert* the desired genetic trait into a new genome (for example a bacterial plasmid).
- *Replication* of the plasmid in a bacterium to have more copies of the gene to be transferred.
- *Transfer* of plasmid in a plant species.

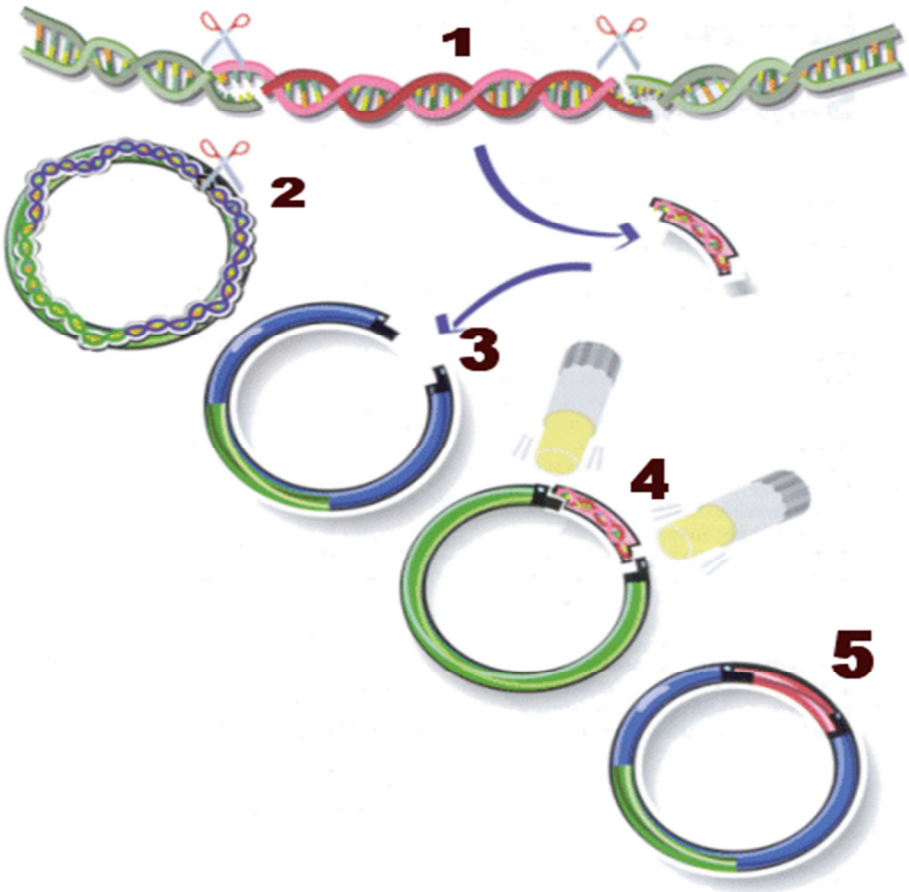


**Figure 2.** Gene Transfer Methods Applicable to Agricultural Organisms

The isolation is carried out by the restriction enzymes which are protein complexes found in some bacterial strains and are able to cut the DNA at specific sequences of bases, different for each enzyme, allowing it to fragment the genome in a precise and reproducible. The cuts may take place within the chain, through the work of the endonuclease, or ends for the work of exonuclease. The use of these enzymes has allowed to isolate the individual DNA fragments. Researchers have identified and use different restriction enzymes, each of which cuts both strands of the double helix in correspondence with a specific sequence of base pairs. The cuts are often staggered so that forming fragments provided with single-stranded ends, said ends sticky or cohesive. Since cutting is always performed at the same sequence of bases for any DNA molecule, irrespective of its origin (bacterial, viral, eukaryotic), all the fragments have complementary cohesive ends, that combine spontaneously.

The techniques that allow the insertion of a gene or a plasmid in a bacterial cell, plant, animal, or in a protoplast (ie a private cell wall and / or cell membrane) are varied; some of them can be used for any cell type while others are specific. The most common techniques are :

- The polymerase chain reaction (PCR)
- Electroporation

- Biolistics
- Calcium Phosphate Precipitation

➢ **The polymerase chain reaction (PCR)**

The Polymerase Chain Reaction (PCR) [7] is now a very widely used technique for copying DNA. Starting with only a small sample of DNA, PCR can generate many copies of a specific DNA segment to be used for further analysis. PCR, requires only the knowledge of the adjacent sequences (primers) to the fragment to be amplified and a polymerase resistant to heat. A typical PCR cycle involves four main stages that are repeated several times:

- Template DNA - the starting DNA of interest
- Two Primers (oligonucleotides) - short, single-stranded, synthesized pieces of DNA that complement sequences on each side of the region of the template DNA that is being amplified
- Thermostable DNA Polymerase - typically Taq (Thermus aquaticus), a heat stable enzyme capable of adding nucleotides to a growing DNA strand
- dNTPs - a supply of the 4 nucleotides needed to make the new DNA strands
- Cationic Magnesium - a cofactor for the polymerase
- Appropriately Buffered Solution - to maintain the pH and salt concentrations appropriate for the polymerase
-

➢ **Electroporation**

The prepped target cells are saturated in a solution with the chosen DNA. A brief but strong electric shock is transmitted through the solution [8], causing little tears in the walls of the cells. This allows for the new genetic material to penetrate the nuclei. Afterwards, the cells are put in a different solution that coaxes the repair of their walls, which works to 'trap' the DNA of the donor in the cell. The chosen DNA becomes joined with the host chromosomes to give the host this new gene.

➢ **Biolistics**

This technique uses the chosen DNA to attach it to tiny gold particles. The particles – now 'carrying' DNA – are forced into the target cells using an intense burst of gas.

➢ **Calcium Phosphate Precipitation**

In this biotechnology technique, the chosen DNA would be exposed to calcium phosphate, which results in the creation of miniscule granules. The targeted cells react to the granules by essentially 'swarming' them and ingesting them, thereby facilitating the granule release of DNA and the subsequent delivery to the host's nuclei and chromosomes.

The techniques through which you can obtain genetically modified organisms are relatively recent and currently available on the market GMOs that have characters that can be controlled with relative ease; in fact they are inserted one or a few genes related to a certain characteristic (a typical example is the resistance to a particular disease); but it must take into account that, over the last decade, genomics has made great strides and it is very likely that we will see soon appear on the market GMOs particularly complex genetic modifications.

## USEFULNESS OF GM CROPS

Genetically modified (GM) foods have an interesting history and their development has experienced rapid growth over the last decade. Within all of the history, there has been a great deal of controversy and debate about the benefits and risks of GM foods and the production process. Given the current situation around GM foods, the controversy will likely continue for some time as well.

Soya - soya has been genetically modified to increase the amount of the C4 protein, a protein that gives the plant increased resistance to herbicides.

Corn - There are some varieties of genetically modified corn.

- One of these, containing the Bacillus thuringiensis (Bt) genetic material useful in the production of a BT toxin capable of poisoning pests of the family insects of lepidoptera (moths and butterflies). Often these varieties of maize are also resistant to several types of herbicides. The corn is hidden in many prepared foods and packaged under the name of cornstarch, glucose and fructose. Be very careful.

Sugar beets - genetically modified to resist the herbicide used by the US giant Monsanto.

Rice - the genetically modified to resist herbicides should not yet be available for human consumption, but some amount of rice (long grain) genetically modified (LLRICE601) [9] have been included in both the US and Europe. It has recently created a new rice strain, golden rice, to increase the natural production of beta carotene that our body uses to make vitamin A. Golden rice is still in the testing phase and has not yet been determined if the 'the human intake can be dangerous or not.

In the agricultural field they have been developed that bacteria introduced in the soil improve its characteristics and are able to protect them from frost or insects. They then obtained more resistant plants to various stress, bacteria or viruses, or more tolerant to certain herbicides.

Where food enzymes are products used for industrial production and for fermentation processes; They are then obtained from plants much better organoleptic characteristics and animal products whose nutritional and organoleptic characteristics are well above the norm.

## HARMFULNESS OF GM CROPS

In the face of the indisputable benefits that could result from the release of GMOs into the environment, they are also evaluate the risks.

Among the major risks cited are those resulting from food safety, which could be jeopardized by the introduction of toxic elements; Furthermore, they may of arising allergies and resistance to drugs in pathogenic organisms. In a study in the early 1990's rats were fed genetically modified (GM) tomatoes. Well actually, the rats refused to eat them. They were force-fed. Several of the rats developed stomach lesions and seven out of forty died within two weeks [6]. Scientists at the FDA who reviewed the study agreed that it did not provide a "demonstration of reasonable certainty of no harm." In fact, agency scientists warned that GM foods in general might create unpredicted allergies, toxins, antibiotic resistant diseases, and nutritional problems. Not insignificant is also the risks arising from the interaction with other organisms, which could give rise to a transfer of genes, the pollution of the genetic basis through the dispersion of seeds or pollen, the transfer of genes in microorganisms (DNA uptake) and finally the generation of new viruses by genetic recombination. GM-fed animals had problems with their growth, organ development and immune responsiveness, blood and liver cell formation, as well as damaged organs, sterility. Risks are increased by the fact that the genes inserted into GM food not only survive digestion, but transfer into body organs and circulation. Transgenes have been found in the blood, liver, spleen and kidneys. DNA can even travel via the placenta into the unborn. According to a July 27, 2004 report from the US National Academy of Sciences (NAS), [7] the current system of blanket approval of GM foods by the FDA might not detect "unintended changes in the composition of the food." The process of gene insertion, according to the NAS, could damage the host's DNA with unpredicted consequences. The Indian Council of Medical Research (ICMR), which released its findings a few days earlier, identified a long list of potentially dangerous side effects from GM foods that are not being evaluated. The ICMR called for a complete overhaul of existing regulations [8]. The only human feeding study ever conducted showed that the gene inserted into soybeans spontaneously transferred out of food and into the DNA of gut bacteria [9]. This has several serious implications. First, it means that the bacteria inside our intestines, newly equipped with this foreign gene, may create the novel protein inside of us. If it is allergenic or toxic, it may affect us for the long term, even if we give up eating GM soy. The same study verified that the promoter, which scientists attach to the inserted gene to permanently switch it on, also transferred to gut bacteria. Research on this promoter suggests that it might unintentionally switch on other genes in the DNA – permanently [10]. This could create an overproduction of allergens, toxins, carcinogens, or antinutrients. Scientists also theorize that the promoter might switch on dormant viruses embedded in the DNA or generate mutations [11]. But in a worldwide there are more than 114 million hectares of crops genetically modified plants, more than half of which are located in the US (51%) while a good 87% of them in the Americas. 99% of the crop is concentrated in a few countries: the US, Canada, South America (Argentina, Brazil and Paraguay), India, China and South Africa. The rice art, crafted from rice plants, shows a map of Italy booting out the GMO (genetically modified organisms) logo. Greenpeace, responsible for the art, is asking the Italian government to keep the country's rice GMO free and respect the rights for the Italian citizen to be able to have food choice **Figure 3.**

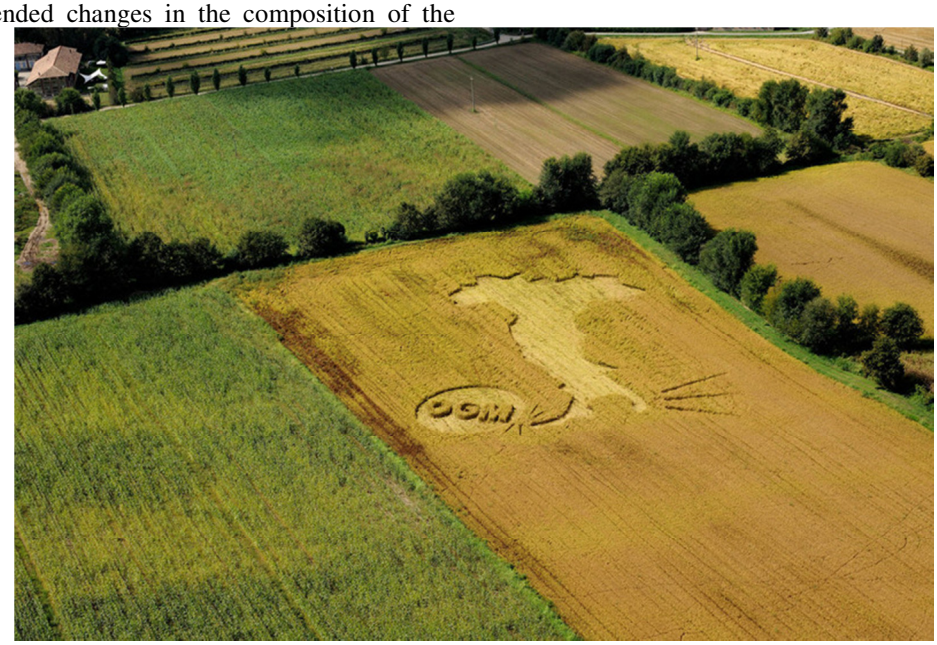


**Figure 3.** Sustainable agriculture rice art in Italy

Applicants can apply for GMO authorizations by submitting a dossier with experimental data and a risk assessment. In 1997, FDA established a voluntary consultation process with GM crop developers to review the determination of "substantial equivalence" before the crop is marketed, such as assessing the toxicity and allergenicity of the gene product and the plant itself. If the data in the food-safety assessment are satisfactory, FDA notifies the developer that marketing of the crop may proceed. Critics have raised questions about whether this voluntary consultation process provides adequate assurance that GM crops are safe. In particular, the use of food crops like corn for the production of non-food products, such as pharmaceuticals, does not fall under FDA's authority unless the gene product ends up contaminating a food crop, at which time the crop is considered adulterated and must be recalled. Because of this gap in regulatory authority, FDA may not perform appropriate oversight until it is too late [12].

In 2004, Senator Dick Durbin (D-IL) introduced legislation that would have required any product grown in a food crop to receive pre-market approval, whether or not it was intended to be eaten. Thus, before any pharmaceutical was produced in a food crop, FDA's Center for Food Safety and Applied Nutrition would conduct a food-safety analysis to ensure that accidental human exposure to the drug through the food supply will not cause health risks [13].
In many countries of the world there are various reference standards on GMOs; such regulations have the purpose of ensuring the greatest possible safety, both in environmental level and at the level of human health and animals. At international level, the relevant legislation is the so-called Cartagena Protocol.

As for the European continent, the texts governing the matter GMOs are as follows:

- Regulation (EC) 1829/2003 on GM food & feed
- Directive 2001/18/EC on deliberate release into the environment
- Commission Implementing Regulation (EU) 503/2013 on applications for authorisation of genetically modified food and feed in accordance with Regulation (EC) 1829/2003 Directive (EU) 2015/412 amending Directive 2001/18/EC as regards the possibility for the Member States to restrict or prohibit the cultivation of GMOs in their territory

## VIETNAM AND GM CROPS

Returning again to the largest factory Monsato and her capacity to produce herbicides that can also kill people. The herbicide who us use is so powerful that the US military uses it as a defoliant in its war in Vietnam, where he conceived the insane idea that destroying all the leaves of the trees of North and Central Vietnam will be able to flush out the Vietcong. Instead they come to Saigon, and they'll run away the American ambassador from the embassy roof, with the Stars and Stripes rolled up under his arm, as he gets up on a helicopter that will bring him back away, forever. Monsanto [14], during all that disastrous war, the first that the Americans lost in their history, the army has sold the infamous "Agent Orange", a mixture of 245T Monsanto and 24D of its rival Dow Chemical, its ally for patriotic destruction of forests in Vietnam. Scientists and the public, in addition to the mass desertions of young Americans do suspend, in 1971, shedding the agent orange, of which there are the effects of dioxins on the environment. It is carcinogenic, has caused immune damage and reproduction that have not finished doing evil to the Vietnamese. The Vietnamese government has never officially stated its stance on the grievous actions of Monsanto and other military contractors for the U.S., focusing instead on reparations for victims of Agent Orange. As one of the makers of Agent Orange, Monsanto claims they were just following the recipe for the formula as directed by the U.S. government. Furthermore, dioxin, found in Agent Orange, is one of the most dangerous chemicals ever made by man. Babies are still being born today with horrific birth defects-decades after Agent Orange was sprayed so haphazardly across Vietnam. Nearly 4.8 million Vietnamese people have been exposed, causing 400,000 deaths and a grab bag of health issues that would make a haunted house seem cheery. An estimated 650,000 victims are suffering from chronic illnesses linked to Agent Orange in Vietnam, alone. Fifty-five years after rendering almost an entire country cancerous, chemical companies like Monsanto are welcomed with open arms into Vietnam. It boggles the mind.

## CONCLUSION

Genetically modified foods have the potential to be many different things but their use and support vary throughout Britain and the rest of the world. One of the most important reasons to think about genetically modified foods is that their production and consumption can affect you-the consumer.

By becoming informed and educated, you can consider all of the different aspects of this issue, which will allow you to make the best political and personal choices regarding the role that genetically modified foods will play in your life. Given that the biotechnology industry is a booming one with enormous annual profits, qualified biotechnology workers are important to ensuring that the industry continues to flourish. You can work in areas such as health care, food production and genetic modification or you may even choose to work in areas that involve an environmental focus. Whether you want to work in the development of genetically modified seeds or you are more keen to work in the laboratory testing and safety aspects of genetically modified products, there is an area in biotechnology that might complement your unique skills, interests and abilities.

Your best approach is to speak to an academic advisor at your school or a local university about how to obtain a career in biotechnology. A typical day in the field of biotechnology can vary a great deal, depending on what subfield you have chosen for your career and employment. Those who work in the laboratory will likely work with a wide range of apparatus. Techniques may include DNA separation or DNA analysis and similar biological techniques.

You may also have a role in the maintenance of organisms used for genetic engineering purposes.

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