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Review

Genetically Modified Plants in Agriculture

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Simple Summary

Genetically modified plants contain an insertion, deletion or edited version of genes of interest within their genomes. A trait gene may originate from an unrelated plant or a different species and can confer specific characteristics (e.g., pest resistance, drought tolerance, salinity tolerance, or resistance to extreme temperatures) or enable the production of proteins with industrial or pharmaceutical value. The first genetically modified plant, tobacco, was produced in the early 1980s. Since then, many plants with a variety of desired traits have been developed. This review outlines the main methods for producing genetically modified plants, lists trait genes employed in agricultural biotechnology, and presents the advantages and disadvantages of utilizing genetically modified plants in agriculture.

Abstract

Genetically modified (GM) plants have revolutionized agriculture for more than three decades. The production of a GM plants is a complex, multi-stage process. Several key methods are available for generating GM plants. The choice of transformation method depends on the type of plant (dicotyledonous or monocotyledonous), the objective (large-scale production versus studying a specific gene in particular cells or tissues), and whether stable or transient transformation is desired. Following successful transformation, the next step is the regeneration of a whole plant from a single cell in tissue culture, which is a labor-intensive and time-consuming process. Currently, numerous genes that confer desirable traits have been identified. These traits include stress tolerance, herbicide and pest resistance, and improved consumer qualities (such as flavor, appearance, shelf life, and nutritional value). In this review, we describe the main methods for producing GM plants and provide examples of trait genes utilized in agricultural biotechnology. Despite the fact that GM plants represent one of the most significant biotechnological advances, they also remain among the most contentious issues in contemporary food safety and agricultural policy. Here, we discuss the advantages and disadvantages of using GM plants for humans.

Keywords: genetically modified (GM) plants; *Agrobacterium*; transformation; transgenes; agriculture; breeding

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1. Introduction

Plant gene technology involves transferring genes with known functional traits, such as high yield potential, stress resistance, disease tolerance, and enhanced nutritional profiles, into target plants or editing or removing specific genes from the plant genome using

advanced scientific methodologies. This process results in the acquisition of novel functional properties while preserving the original genetic foundations of the plant [1,2]. The first genetically modified (GM) plant was produced in the early 1980s by incorporating the *neomycin phosphotransferase II* (*nptII*) gene, which confers resistance to the antibiotics kanamycin and neomycin, into tobacco [3]. Since that time, a multitude of GM plants with various desirable traits have been developed. There are several basic methods for producing GM plants; however, the majority have been generated using the biolistic method or through the use of *Agrobacterium*. Transgenes may originate from an unrelated plant or from an entirely different species and can endow the plant with specific traits or enable the production of proteins with industrial and pharmaceutical value [2].

In this review, we outline the main methods for producing GM plants, compare traditional and molecular plant breeding, and provide some examples of trait genes used in agriculture. Additionally, we discuss the advantages and disadvantages of using GM plants in the context of their impact on human health.

2. Transformation Methods

The hallmark of GM technology is its ability to transfer genes across species, thereby enabling the introduction of desirable traits that may not be naturally present in the target organism. These genes can be sourced from the same or closely related species, or from different species entirely, which expands the genetic resources available for crop improvement [4] compared to those available through traditional techniques [5]. The choice of methodology for obtaining a GM plant is determined by several factors: (1) whether the plant belongs to the class of dicotyledons or monocotyledons. For most dicotyledons, *Agrobacterium*-mediated transformation is preferred, whereas, in the case of monocotyledons, biolistics is more commonly employed; (2) the type of target tissue. Various tissues and organs can be subject to transformation, including leaf pieces (discs), immature embryos, callus cells, and pollen; and (3) the research objectives. To study transient gene expression, simple delivery methods for individual cells are suitable. To obtain a stable inheritable line, gene integration into the genome followed by the regeneration of the plant is required. Each method has its own advantages and limitations [6], and the selection of the optimal approach is crucial for successfully creating a transgenic plant with the desired traits—such as resistance to herbicides, pests, or abiotic stresses, or improvements in nutritional quality. All methods of plant transformation can be divided into two main types: indirect (also known as vector-mediated gene transfer) and direct gene transfer of foreign DNA into the plant cell (Figure 1).

2.1. Vector-Mediated Gene Transfer

2.1.1. "*Agrobacterium*" Method

This method is based on the utilization of *Agrobacterium* and the ability of these bacteria to transfer their genes into the plant genome. The species most commonly used for transformation is *A. tumefaciens*, which contains the tumor-inducing (Ti) plasmid responsible for causing crown gall disease. A key component of the Ti plasmid is the region of transfer DNA (T-DNA), which encodes the biosynthesis of opines and phytohormones. The three oncogenes (involved in the biosynthesis of opines, cytokinins, and auxins) located within the T-DNA are the primary causes of tumor formation in plants. Plant transformation employs modified agrobacteria that lack tumor-promoting genes or opine-synthesis genes in their genome. A vector is created based on the Ti plasmid, from which these native genes are removed to prevent harm to the plant. In their place, the desired trait gene, a selection marker, and optionally a reporter gene for visual confirmation of successful transformation are inserted.

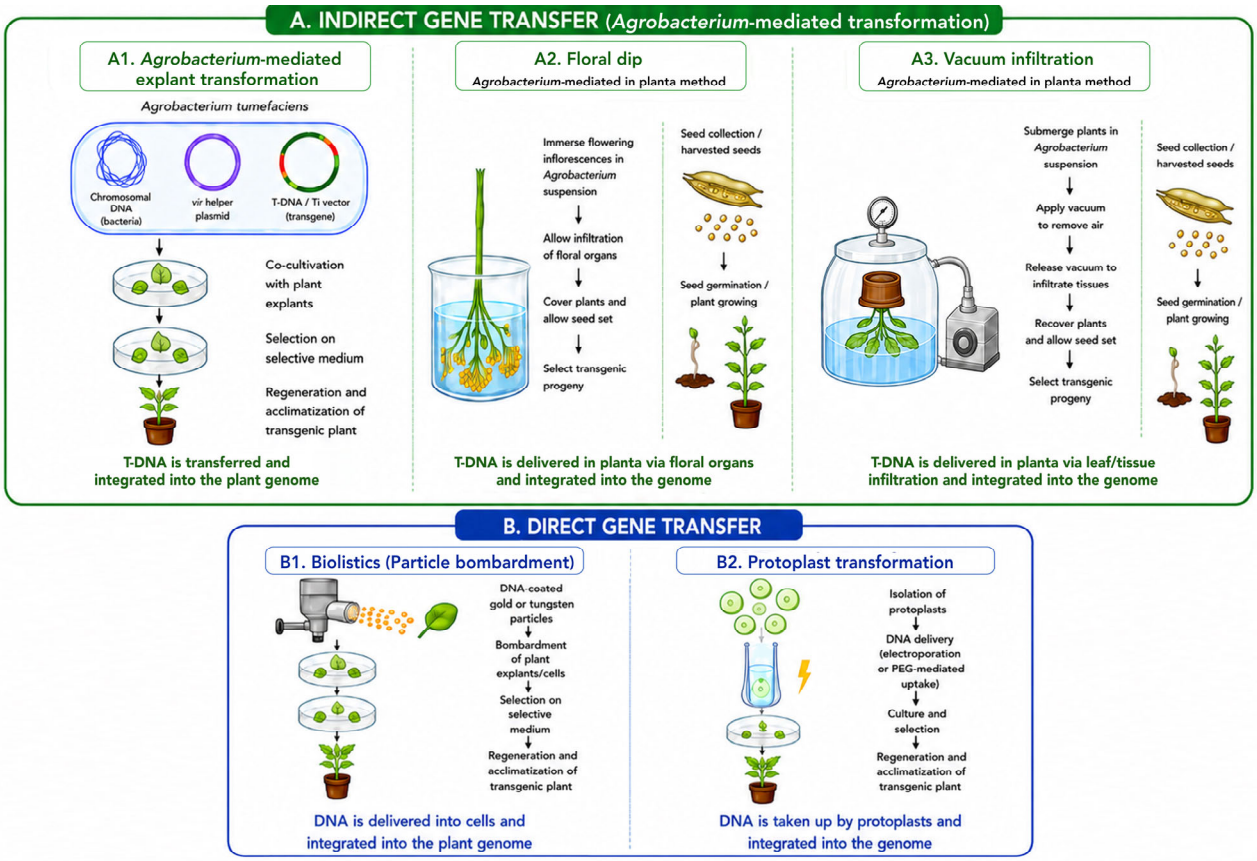


Figure 1. Indirect and direct DNA-transfer methods in plant transformation.

There are two main types of vectors used for *Agrobacterium*-mediated plant transformation: cointegrate vectors and binary vectors [7]. The strategy based on cointegrate vectors is not widely employed today due to the complex engineering required for these vectors. A more commonly utilized and convenient system for *Agrobacterium*-mediated transformation is the T-binary vector system, which comprises two components: the T-binary vector and the *vir* helper plasmid (Figure 1A) [8–10]. The T-binary vector contains T-DNA border repeats derived from the Ti plasmid as well as the genes of interest. It can replicate independently in both *E. coli* and agrobacteria, separate from the bacterial chromosome. After the production and purification of the T-binary vector from *E. coli*, it is transformed into agrobacteria, such as strains LBA4404, EHA105, AGL-1, or GV3101 [11–13]. These strains harbor the *vir* helper plasmid, which is a disarmed Ti plasmid devoid of tumor-related genes, responsible for the synthesis of Vir proteins [8,14]. Agrobacteria containing the T-binary vector are cultivated in a nutrient medium supplemented with appropriate antibiotics to ensure an adequate number of cells for plant infection. For the transformation process, plant tissues or explants exhibiting a wounded surface are employed. During the co-cultivation of these damaged plant parts with a suspension of agrobacteria, plant cells release a specific phenolic compound (acetosyringone), which activates *vir* genes in the agrobacteria [15–18]. The products of the *vir* genes, along with certain proteins, facilitate the excision and transfer of single-stranded T-DNA from the T-binary vector into the host plant cell [19]. Within the nucleus, the T-DNA is integrated into the plant genome with the assistance of Vir proteins. Subsequently, the explants are transferred to a nutrient medium containing antibiotics to eliminate the agrobacteria and to select for transformed cells.

The subsequent step, the regeneration of a whole, fertile plant from a transformed cell in tissue culture, represents a critical phase [20,21]. This developmental process predominantly occurs through two distinct pathways: indirect organogenesis and somatic

embryogenesis. In the case of indirect organogenesis, dedifferentiation is initiated by a precise balance of exogenous auxins and cytokinins on the surface of the wounded explant, resulting in the formation of a callus—an unorganized mass of cells. Subsequently, shoot apical meristems are induced on a selective medium characterized by a high cytokinin-to-auxin ratio, followed by root induction on an auxin-enriched medium [22]. During somatic embryogenesis, transformed cells undergo a fate transition and dedifferentiation to acquire embryogenic competence. These cells then develop into bipolar somatic embryos, either completely bypassing the callus phase or transitioning through a brief, transient callus stage [23].

In vitro regeneration remains the most labor-intensive step and acts as the primary bottleneck (recalcitrance) in generating transgenic lines (Table 1) [24]. This limitation is particularly pronounced in monocotyledonous crops, where regeneration efficiencies remain strictly dependent on specific, hard-to-obtain explants, severely restricting high-throughput genetic engineering and genome editing [25].

Table 1. Advantages and limitations of DNA-transfer methods in plant transformation.

Method	Main Advantages	Key Limitations
Agrobacterium-mediated transformation	requires minimal equipment and relatively straightforward operation; low transgene copy number; few DNA rearrangements and comparatively stable transgene expression	many monocots, legumes and woody species remain recalcitrant, because of restricted host range; plant regeneration is highly time-consuming
plant virus vectors	high-level transient expression without genome integration; suitable for VIGS, VIGE, genome-editing reagent delivery and recombinant protein production	limited cargo capacity and genetic instability of many (+)ssRNA virus vectors; restricted meristem/germ cells delivery in some systems; narrow host range or difficulty carrying foreign DNA fragments in certain DNA virus vectors; small insert capacity of Caulimovirus-based particles
biolistics	applicability to both dicotyledonous and monocotyledonous plants, including *Agrobacterium*-recalcitrant cereals and legumes; stable and transient expression; simultaneous delivery of multiple genetic elements; organelle transformation, including chloroplast transformation	multiple-copy insertion; rearranged transgenes; integration at multiple genomic locations; off-target deletions; possible tissue damage or compromised DNA integrity during delivery
protoplast transformation	simple and reproducible workflow; suitability for transient expression assays, gene function studies, protein localization and preliminary CRISPR/Cas9 editing screens	restriction mainly to protoplasts or a small number of cell types; species-, organ- and tissue-dependent isolation conditions; difficult and time-consuming regeneration of healthy plants from protoplasts
microinjection	precise delivery into selected cells or protoplasts; direct targeting of cytoplasm or nucleus; independence of the type of plant	labor-intensive and technically demanding procedure; low throughput; requirement for costly specialized equipment, trained personnel and efficient recovery or regeneration systems
nanotechnology-based delivery	non-invasive and diffusive delivery properties; potential alternative for *Agrobacterium*-resistant species; targeted delivery; low toxicity; cargo protection; compatibility with transient and stable genetic modification	emerging technological status; recent development of nanotechnology-based plant gene delivery; insufficient exploration of efficient nanoparticle delivery into plants

At present, numerous protocols feature various modifications of the plant transformation process using *Agrobacterium* [26,27]. These modifications include the optimization of the nutrient medium composition, bacterial strains, types of explants, and the use of hormones and antibiotics. Moreover, superbinary vectors have emerged as an improved version of binary vectors, carrying additional *vir* genes responsible for the supervirulence phenotype of the bacteria [28,29]. The superbinary vector-based system exhibits a remarkably high frequency of transformation, which is particularly valuable for recalcitrant plants such as cereals [28]. In addition, in planta transformation protocols have emerged, significantly increasing transformation efficiency and allowing for the successful modification of both dicotyledonous and monocotyledonous plants, including major cereal crops such as wheat, rice, and maize [30–34].

The two most common *Agrobacterium*-mediated in planta methods are floral dip and vacuum infiltration (Figure 1A) [35]. The floral dip method involves immersing flowers in an *Agrobacterium* suspension [36,37]. The resulting transgenic seeds are collected directly from the plant. While this approach is quick and practical, it exhibits low transformation efficiency and is primarily suitable for dicotyledonous plants [36,38–41] and some monocot plants [42,43].

Vacuum infiltration, where plant tissue or whole plants are submerged in a liquid suspension of *A. tumefaciens* and subjected to reduced pressure followed by rapid repressurisation, is a common method for introducing bacteria into plant tissue (Figure 1A) [33,44]. The vacuum infiltration method allows for the production of both transgenic seeds and transgenic vegetative parts of plants. This approach has long been employed to increase transformation efficiency in numerous plants, including both dicotyledonous and monocotyledonous species, as it enhances the penetration of agrobacteria into the layers of plant tissue [45,46]. Furthermore, the vacuum infiltration method can be complemented by sonication, resulting in even higher transformation efficiency [47–51].

2.1.2. Methods Based on Plant Virus Vectors

Plant virus vectors are employed as tools for the effective and precise delivery of genetic material into plants [52,53]. RNA and DNA viruses differ fundamentally in their infection mechanisms and are therefore utilized to deliver distinct types of molecular cargo [54]. The most common plant viruses contain single-stranded RNA (ssRNA) and are classified into two types: positive-strand ((+)ssRNA) and negative-strand ((−)ssRNA) viruses [54].

The majority of (+)ssRNA viruses can only accommodate insertions of a few hundred nucleotides (Table 1) [55–57]. Due to their limited cargo capacity and genetic instability, (+)ssRNA viruses are unable to deliver long foreign sequences (for instance, the Cas9 open reading frame [58]), but they are well suited for smaller cargoes such as guide RNAs.

(−)ssRNA viruses exhibit greater genome stability and higher cargo capacity compared with (+)ssRNA viruses [58–61]. They are capable of accommodating an entire CRISPR/Cas cassette, thereby eliminating the need for a Cas nuclease-expressing transgenic recipient plant. However, (−)ssRNA viruses are excluded from the meristem and cannot deliver editing reagents into germ cells [62]. Consequently, edited somatic cells must be regenerated into whole plants that can transmit the genetic modifications to their progeny [54].

Plant DNA viruses are less abundant compared with plant RNA viruses. Among them are the single-stranded DNA (ssDNA) viruses belonging to the families *Geminiviridae* [63,64] and *Nanoviridae* [65], as well as the double-stranded DNA (dsDNA) plant pararetroviruses of the family *Caulimoviridae* [66,67].

Geminiviruses have facilitated the development of virus vector systems capable of delivering long donor DNA fragments and achieving high copy numbers [68,69]. Additionally, Geminiviruses have been utilized as vectors for virus-induced gene silencing (VIGS), which degrades transcripts of endogenous plant genes without altering the genes themselves or necessitating stable genetic transformation (Table 1). Furthermore, Geminivirus-based vectors offer a robust platform for delivering genome editing reagents. Virus-induced genome editing (VIGE) is a technique that employs engineered plant viruses to transport CRISPR/Cas components into plant cells. Typically, the viral genome is encoded within the T-DNA borders of a binary vector, and the virus is assembled upon delivery to the plant cell. Due to its high replication rate, the CRISPR/Cas components are expressed at elevated levels [70].

Members of the family *Caulimoviridae* have primarily been utilized as sources of regulatory elements [71]. The most notable example is the 35S promoter from Cauliflower mosaic virus (CaMV), which remains one of the strongest constitutive promoters available for use in plants [72]. However, their application as replicating vectors is limited by a narrow host range and difficulties associated with the insertion of foreign DNA fragments. The size of exogenous DNA that can be successfully propagated within viral particles is restricted to approximately 250 bp [73,74].

Several techniques are available for delivering viral vectors into plants, including mechanical inoculation [75–77], foliar spraying [78,79], needle-laden injection into the stem or petiole [78], agroinfiltration [55,80], and the biolistic method [54,78].

2.2. Direct Gene Transfer

2.2.1. Biolistic Method

The 'Gene Gun', also known as the particle bombardment method or biolistics, is one of the most widely used techniques for plant transformation [81,82]. This approach is particularly useful for plant species that are difficult to transform using *Agrobacterium*-mediated methods, including many cereals and legumes [6]. The name of the technique derives from the process by which cells are 'shot' with genetic material. In practice, the procedure begins by mixing the DNA of interest with tiny particles made of gold or tungsten. These metal particles carry a positive charge, which binds them to the negatively charged DNA. Once coated, the DNA-metal particles are loaded into a gene gun or biolistic device. A pressurized gas, typically helium, then propels a microcarrier bearing the DNA-coated particles towards a stopping screen. As the gas pressure increases, the microcarrier forces the DNA-metal particles through the screen, allowing them to penetrate the cell membranes and deliver the DNA constructs directly into the nucleus of target cells placed in a Petri dish (Figure 1B) [83]. Following delivery, the DNA detaches from the metal particles and may be integrated into the plant's chromosomal DNA by endogenous recombination mechanisms.

Biolistics is effective for transforming both dicotyledonous and monocotyledonous plants (Table 1). Similar to *Agrobacterium*-mediated transformation, it can achieve both stable and transient gene expression, as well as the simultaneous delivery of large numbers of different genetic elements [84]. The technique is relatively non-toxic and can be used to introduce DNA into almost any type of tissue, including immature and mature embryos, shoot apical meristems, leaves, roots, and others [85]. However, this method has several disadvantages, which are detailed in Table 1.

2.2.2. Transformation of Protoplasts

Protoplast transformation is a technique used to introduce foreign DNA into plant cells that lack a cell wall, known as protoplasts or naked cells. This approach is commonly employed to study gene function, determine protein localization, or perform CRISPR/Cas9

genome editing. It is particularly useful for transient gene expression, allowing the analysis of multiple genes within a short timeframe (Table 1).

In practice, protoplasts are first generated through enzymatic digestion that breaks down the cell wall. Notably, protoplasts largely retain the cellular identity and differentiated characteristics of their original source cells [86]. Consequently, the isolation procedure must be tailored to each species, organ, or tissue [87]. Following isolation, direct DNA delivery into individual plant cells is achieved using either polyethylene glycol (PEG) or electroporation (Figure 1B) [88–90]. PEG-mediated transformation is an effective and widely adopted method for directly delivering DNA into protoplasts. In the presence of calcium ions, PEG induces reversible membrane fusion and promotes the uptake of exogenous DNA via endocytosis [91]. Electroporation-based protoplast transformation uses electrical pulses to generate transient pores in the plant cell membrane, thereby facilitating the entry of foreign DNA [92]. After transformation, protoplasts are selectively cultured and can be used for subsequent regeneration steps [93].

2.2.3. Microinjection

The microinjection technique represents a direct physical strategy for delivering DNA into selected cells [94,95]. It employs a fine glass needle operated under microscopic observation, allowing precise injection without harming the target cells. This method is labor-intensive and requires both expensive equipment and considerable technical skill. In practice, the target cell is held steady beneath a microscope using two micromanipulators. One of these functions as a holding pipette to secure the cell in place, while the other is a microcapillary tube filled with a small volume of DNA solution, intended to pierce either the plasma membrane or the nuclear envelope. Through this process, DNA is introduced into the cytoplasm or nucleus of plant cells or protoplasts using a microcapillary pipette. Following the completion of gene transfer, the transformed cells are cultured and eventually regenerated into whole plants. This technique can be applied both to individual cells and to protoplasts.

2.2.4. Nanotechnologies

Nanotechnologies facilitate precise genetic transformation in plants by circumventing the rigid plant cell wall through the use of nanoparticles, which are ultrafine particles with diameters of less than 100 nm. Various types of nanoparticles, including carbon dots, as well as single-walled and multi-walled carbon nanotubes, gold nanoparticles (nanospheres, nanorods, nanoclusters), silicon-based carriers, magnetic nanoparticles, chitosan nanoparticles, and bio-inspired carriers such as liposomes and vesicles, are capable of transporting different types of cargo, including drugs, proteins, and nucleic acids, into plant cells [96–98]. The plant transformation approach using nanoparticles exploits the similar electrical charge near the cell membrane to overcome the barrier of the plant cell wall, thereby introducing various cargoes into plant tissues to achieve either transient or stable transformation [99]. Additionally, nanoparticles have been successfully employed for organelle-targeted gene delivery [100].

3. Plant Breeding: Traditional Versus Molecular Approaches

Using the aforementioned techniques, genetic engineering is carried out, which involves the precise, direct modification of an organism's DNA using recombinant DNA technology or gene-editing tools like CRISPR. It inserts, removes, or alters specific genes to introduce entirely new and highly targeted traits. Genetic engineering, alongside marker-assisted selection (MAS) and genomic selection, is a component of molecular breeding [101].

Molecular breeding overcomes several limitations of traditional breeding by combining marker- and genome-based selection with targeted genetic modification approaches. DNA markers, genome sequencing, and genomic selection enable breeders to identify desirable genotypes at the molecular level before full phenotypic expression, whereas transgenesis and genome editing allow for the direct modification of genetic variation. By shifting the focus from visible phenotypes to molecular markers, genomic loci, and genotype-phenotype associations, molecular tools can reduce the time, space, and resources required to identify complex, recessive, or environmentally sensitive traits [102]. This distinction between phenotype-led and genotype-informed breeding is summarized in Figure 2, which contrasts the conventional workflow of crossing, phenotypic selection, and field-testing with molecular approaches based on marker detection, genomic prediction, and targeted genetic modification.

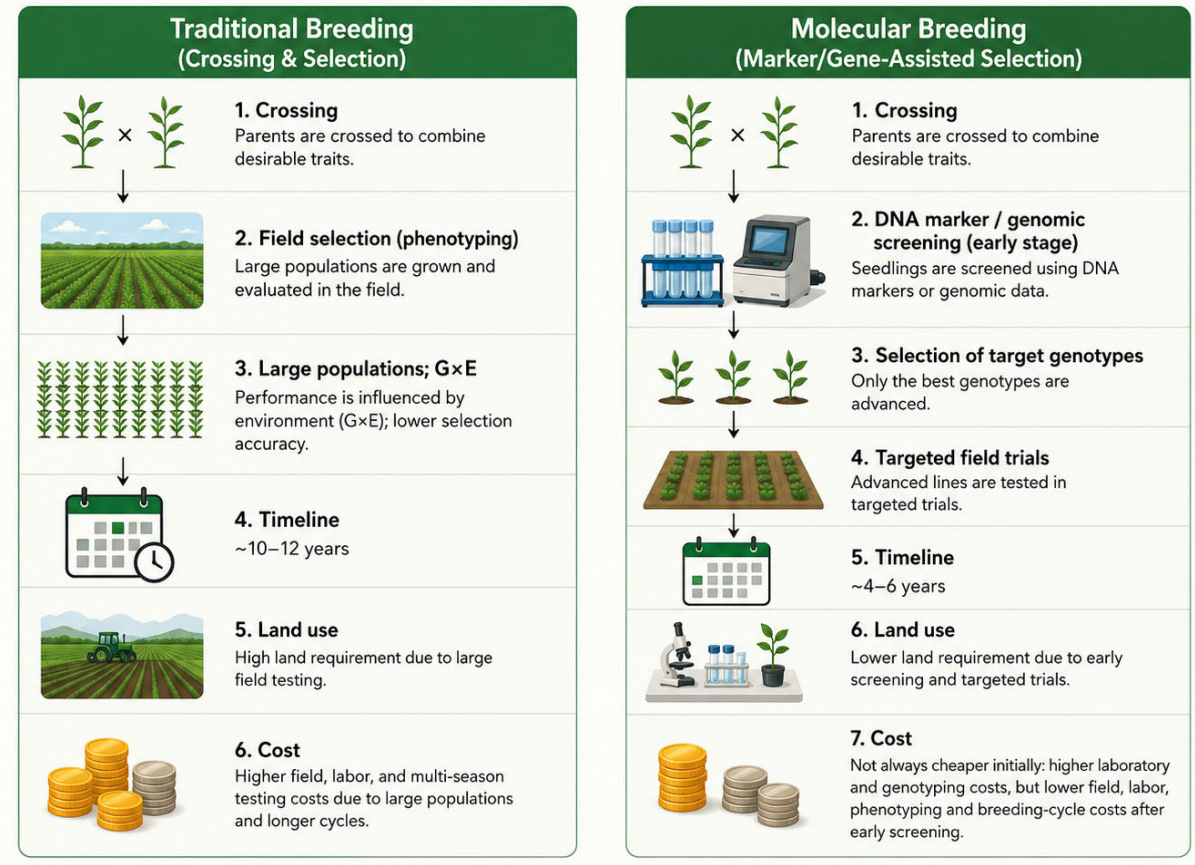


Figure 2. Comparison between traditional and molecular breeding frameworks.

Gene editing, particularly CRISPR/Cas-based systems, extends molecular breeding beyond selection by enabling targeted DNA modification. Unlike conventional GM technologies, which typically introduce transgenes into the host genome, CRISPR/Cas systems can introduce programmable, site-specific changes (Figure 3). In some instances, CRISPR-edited plants can be generated without retaining foreign DNA [5].

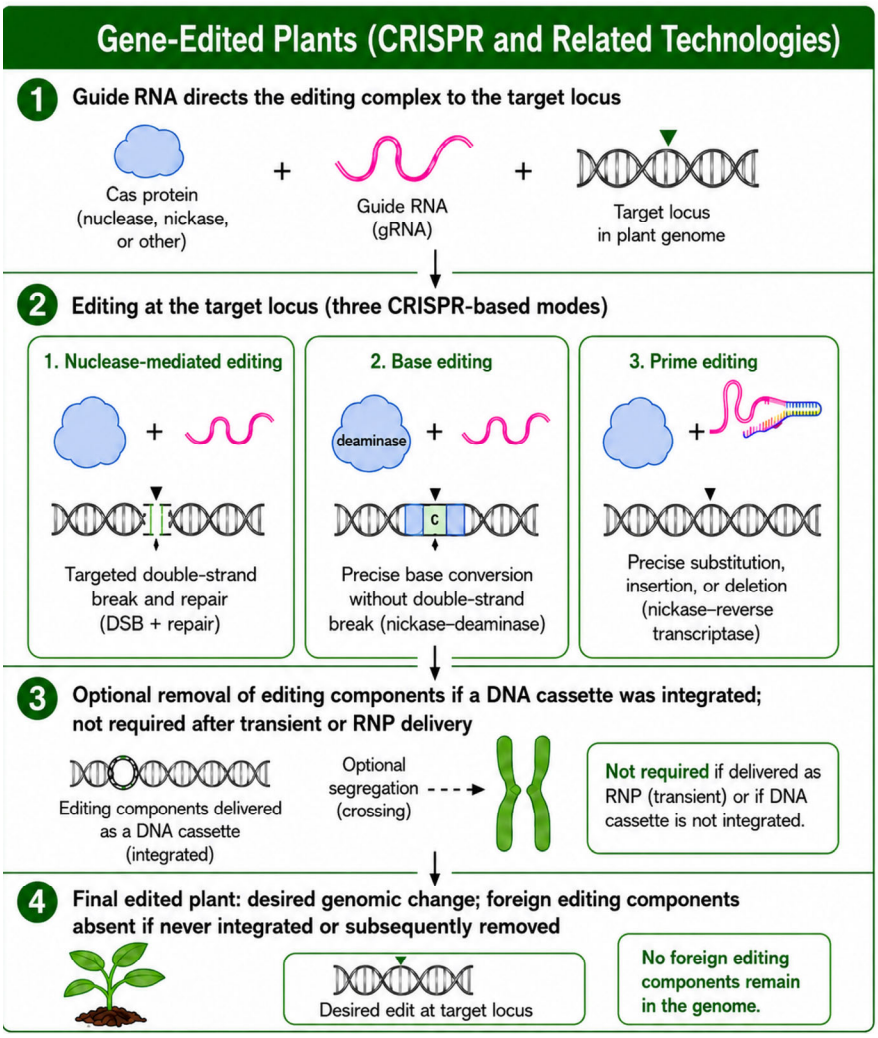


Figure 3. CRISPR/Cas genome editing.

Molecular breeding is also experiencing a temporal shift. Earlier approaches primarily focused on single-nucleotide polymorphisms (SNPs), quantitative trait loci (QTL) mapping, and MAS, whereas modern applications increasingly trend towards broader genome engineering. Recent developments include programmed structural variation, such as large insertions, duplications, inversions, and transposable-element-based genome reshaping [103]. Furthermore, base editing expands CRISPR-based breeding by enabling targeted nucleotide conversion while avoiding the formation of double-strand breaks, making it beneficial for the precise modification of agriculturally relevant alleles [104]. Additionally, CRISPR-based epigenome editing further extends this approach from DNA-sequence alteration to regulatory control. In *Arabidopsis*, directed manipulation of the H3K4me3 chromatin mark has been employed to influence endogenous gene expression, resistance-related responses, and recombination in low-recombining genomic regions [105].

Ultimately, molecular breeding should not be regarded as a substitute for traditional methods. Although molecular tools expedite genetic advancement and improve precision, field validation remains essential, as the ultimate agronomic performance is significantly influenced by environmental conditions and genotype × environment (G×E) interactions (Figure 2). Therefore, the ideal modern breeding paradigm is an integrated one: conventional breeding supplies agronomic and field-testing frameworks, while molecular biology, genome editing, AI-assisted prediction, and tissue-culture-free delivery strategies provide mechanisms for more precise and accelerated selection or modification.

4. Trait Genes

Currently, numerous plants have been artificially created by humans and possess specific transgenes that confer particular traits. In this section, we consider a number of genes used to endow agricultural crops with various beneficial properties, such as herbicide resistance, enhanced biomass, resistance to pests and diseases, improved consumer qualities, stress response, and resistance to adverse environmental factors.

4.1. Genes Providing Herbicide Resistance

Herbicides have a complex and far-reaching impact on plant genes [106]. They can induce random chromosomal changes and mutations or serve as a powerful force of natural selection, eliminating susceptible individuals and promoting the accumulation of plants possessing protective mutations within populations [107]. There are two distinct types of herbicide resistance mechanisms: target-site resistance (TSR) and non-target-site resistance (NTSR) [107,108]. TSR involves mutations within the genes that code for herbicide target proteins [109]. Most of these mutations are non-synonymous substitutions that lead to an amino acid change, thereby preventing the herbicide from binding to the protein without disrupting the enzyme's normal function [110]. TSR may also result from increased expression of the target gene, resulting in the production of more enzyme than can be effectively inhibited by standard herbicide application rates [111,112].

NTSR is a complex and rapidly evolving mechanism that enables weeds to survive following herbicide treatment. Whereas TSR arises from mutations in a specific target protein, NTSR can involve a large number of genes and is achieved by reducing the quantity of active herbicide through sequestration, decreased herbicide absorption, translocation, and enhanced metabolism [113,114]. This form of resistance is particularly concerning because it often confers cross-resistance to herbicides with different modes of action [115]. NTSR presents a significant challenge for agriculture. This resistance mechanism typically evolves as a consequence of prolonged herbicide application, during which various resistance alleles accumulate within a population. Due to its polygenic basis and its ability to provide cross-resistance to new or even yet-to-be-utilized herbicides, weeds possessing NTSR are considerably more difficult to manage than those with TSR.

Examples of genes that confer herbicide resistance are presented in Table 2.

Table 2. Herbicide resistance genes.

GM Plant	Gene	Source	Desirable Trait	Ref.
maize, soybean, cotton, rice, sweet potato, *Arabidopsis*	*4-hydroxyphenylpyruvate dioxygenase* (*HPPD*)	rice, maize, sweet potato, cotton, *Pseudomonas fluorescens*, *Avena sativa*	resistance to HPPD-inhibiting herbicides, increased resistance to abiotic stresses	[116–122]
Eleusine coracana, *Glycine max*, *Linum usitatissimum*, *Nicotiana plumbaginifolia*, *Nicotiana sylvestris*	*alpha*- and *beta-tubulin*	*Eleusine indica*, *Setaria viridis*, *Lolium rigidum*	resistance to herbicides belonging to the group of the microtubule inhibitors	[123,124]
wheat, *Arabidopsis*	*acetolactate synthase* (*ALS*)	*Schoenoplectiella juncoides*, *Triticum aestivum*, *Beckmannia syzigachne*, *Bromus japonicus*, *Echinochloa phyllopogon*, *Schoenoplectiella juncoides*	resistance to ALS inhibitors	[125–129]
soybean, rice, canola	*5-enolpyruvylshikimate-3-phosphate synthase* (*EPSPS*)	rice, *Eleusine indica* (L.) Gaertn., *Conyza canadensis*, *Amaranthus palmeri*, *Amaranthus tuberculatus*, *Amaranthus hybridus*, *Chloris truncata*, *Lolium perenne* ssp. *multiflorum*, *Agrobacterium* sp. strain *CP4*	resistance to glyphosate	[130–135]

Table 2. *Cont.*

GM Plant	Gene	Source	Desirable Trait	Ref.
maize, soybean, canola, cotton	*phosphinothricin acetyltransferase* (*pat*) *bialaphos resistance* (*bar*)	*pat* gene from *Streptomyces viridiochromogenes* *bar* gene from *Streptomyces hygroscopicus*	resistance to glufosinate	[136,137]
canola, alfalfa, cotton, maize, soybean	*glyphosate N-acyltransferase* (*GAT*)	soil microorganisms from extremely glyphosate-polluted soil	resistance to glyphosate	[138–140]
cotton, soybean, maize, *Arabidopsis*	2,4-D degrading enzymes (TfdA, RdpA, SdpA)	TfdA from *Ralstonia eutrophus*, RdpA from *Sphingobium herbicidivorans*, SdpA from *Delftia acidovorans*	resistance to 2,4-dichlorophenoxy acetic acid (2,4-D)	[141–143]
rice, *Agrostis stolonifera* L.	*acetyl-CoA carboxylase* (*ACCase*)	*Alopecurus myosuroides*, *Echinochloa crus-galli* (L.) P. Beauv., *Lolium multiflorum* Lam., rice	resistance to ACCase-inhibiting herbicides	[144–149]
rice, tobacco, *Arabidopsis*	*psbA*	*Raphanus raphanistrum*, *Lolium perenne* L. ssp. multiflorum (Lam.) Husnot, maize, *Arabidopsis*	resistance to PSII-inhibiting herbicides, improved drought tolerance, enhanced sulfur dioxide tolerance, enhanced plant heat tolerance	[150–153]
rice, *Arabidopsis*	F-box proteins	rice, *Arabidopsis*	resistance to synthetic auxin herbicides (picloram, dicamba)	[154,155]
maize, soybean, cotton, canola	*dicamba monooxygenase* (*dmo*)	*Pseudomonas maltophilia* strain DI-6	resistance to the herbicide dicamba	[156,157]

4.2. Genes Responsible for Increasing Biomass

Genetic engineering offers opportunities to significantly enhance the productivity of agricultural crops. Researchers are developing transgenic varieties that exhibit faster growth, produce greater biomass, and possess improved tolerance to environmental stress. These advancements are achieved through the introduction of genes that regulate key physiological processes in plants, ranging from the control of photosynthetic efficiency to the modulation of plant hormonal systems. Such genes include the following: *RPS6K2* [158], *fto* [159], *CYP85A3* [160], *D11-2A* [161], *PTR6* [162], *NFYA-B1* [163], *NLP7* [164], and others. In Table 3, we present some of the genes that enhance biomass.

Table 3. Genes for enhancing biomass.

GM Plant	Gene	Source	Desirable Trait	Ref.
poplar, *Arabidopsis*	*Booster*	poplar	improved photosynthetic efficiency, increase in biomass and seed yield	[165]
rice, *Arabidopsis*, potato, tobacco, tomato	*malate synthase* (*MS*) and *glycolate dehydrogenase* (*GDH*)	*MS* gene from pumpkin and the *GDH* gene from the alga *Chlamydomonas*	reduced photorespiration, increased photosynthetic efficiency, enhanced biomass and yield	[166–169]
tobacco, *Melia azedarach*, poplar, potato, maize, rice, tomato, *Hibiscus cannabinus* L., *Panicum virgatum* L.	*Gibberellic Acid 20 oxidase* (*GA20ox*)	*Arabidopsis*, *Pinus densiflora*, maize, rice	increased level of active gibberellins in plant tissues, leading to faster growth and greater biomass accumulation	[170–173]
wheat, tomato, tobacco	*sedoheptulose-1,7-bisphosphatase* (*SBPase*)	*Brassica napus*, *Brachypodium distachyon*	enhancing the central metabolic process of carbon fixation, increased photosynthetic rates, increased plant biomass and seed yield, increase in starch accumulation, improved resistance to chilling stress	[174,175]
Arabidopsis, tobacco, rice, *Brassica napus*	*DWARF4* (*DWF4*)	*Arabidopsis*, *Echinacea purpurea*	increased seed yield, higher root biomass and root length, tolerance to dehydration and heat stress, resistance to fungal pathogens	[176,177]

Table 3. *Cont.*

GM Plant	Gene	Source	Desirable Trait	Ref.
rice, canola, wheat, barley	*alanine aminotransferase (AlaAT)*	*Hordeum vulgare*, barley	increased above-ground biomass and seed yield, higher root biomass production	[178–180]
Arabidopsis, rice, cotton, maize, alfalfa, wheat, barley, creeping bentgrass	*vacuolar pyrophosphatase1* (*VP1*)	*Arabidopsis*, *Thellungiella halophila*	enhanced biomass in shoot and root systems, improved phloem loading and transport, resistance to salt stress, improved drought resistance	[181–185]

4.3. Genes for Enhancing Resistance to Pests and Diseases

Improving the resistance of crops to pests and diseases is achieved through the use of specific genetic approaches, notably resistance genes, susceptibility gene editing, and RNA interference (RNAi) [186,187]. Key strategies include the introduction of resistance genes, which enable plants to recognize pathogens, and the application of CRISPR/Cas9 to disrupt susceptibility genes that pathogens rely upon [188,189]. The most widely used resistance genes are *Cry* transgenes derived from the soil bacterium *Bacillus thuringiensis* (Bt) [190]. Other genes employed for the protection of plants against pests and diseases include *Vip* [191], *CpTI* [192], *bgn13.1* [193], *NPR1* [194]. Some of the genes are presented in more detail in Table 4.

Table 4. Genes improving resistance to pests and diseases.

GM Plant	Gene	Source	Desirable Trait	Ref.
maize, tobacco, potato	Chitinase genes	*Spodoptera littoralis*, the *Autographa californica* nuclear polyhedrosis virus, *Penicillium ochrochloron* Q-3-1, *Streptomyces griseus* strain HUT6037	resistance against insect pests and fungal diseases	[195–198]
barley, rice	Antimicrobial peptides (AMPs)	*Drosophila melanogaster*, *Hyalophora cecropia*	plant protection from bacteria, fungi and viruses	[199–201]
tomato, alfalfa	human lactoferrin (hLf)	human milk	enhanced resistance to the phytopathogens	[202–205]
maize, cotton, soybean	*Cry* genes	*Bacillus thuringiensis*	resistance to the bollworm, resistance to the western corn rootworm	[206–208]
tobacco, potato	*inhibitor of serine proteases* (*ISP*)	buckwheat	resistance to bacteria, insects and fungal pathogens	[209,210]
rice	*Pi21*	rice	partial resistance to *Magnaporthe oryzae*	[211–213]
tobacco	*thaumatin*	*Thaumatococcus daniellii*	enhanced tolerance to fungal pathogens and abiotic stresses	[214]
rice, *Camellia sinensis* L. (O.) Kuntze	*Osmotin*	rice, tobacco	increased resistance to *Schizotetranychus oryzae*, improved tolerance to drought stress	[215–217]

4.4. Genes Improving Consumer Properties

Transgenes that enhance the consumer properties of a plant encompass genes responsible for improving flavor, appearance, nutritional value, texture, and shelf life. For instance, in tomatoes, numerous genes that regulate fruit coloration, morphology, flavor, and nutritional value have been identified (reviewed in [218]). In rice, modifications have been made to both amino acid and protein composition [219,220], as well as micronutrient

content [221] (reviewed in [222]). In *Zea mays* L. *saccharata*, the combination of the *sh2* and *se1* genes, or the *su1* and *se1* genes, yields superior hybrids with improved taste, shelf stability, and consumer appeal (reviewed in [223]). In Table 5, we present some genes that enhance the consumer properties of crops.

Table 5. Genes enhancing consumer properties.

GM Plant	Gene	Source	Desirable Trait	Ref.
Golden Rice, Golden Rice 2	*phytoene synthase* (*psy*) in combination with *carotene desaturase* (*crtI*)	*psy* from *Narcissus pseudonarcissus* (Golden Rice), *psy* from maize (Golden Rice 2), *crtI* from *Erwinia uredovora*	biosynthesis of provitamin A in the rice endosperm	[224,225]
rice, barley, potato	*granule-bound starch synthase* (*GBSS*) genes	*Fagopyrum tataricum* (L.) Gaertn., barley, rice	altered starch content and structure	[226–228]
poplar, alfalfa	*Cinnamoyl-CoA Reductase* (*CCR*) genes	poplar, alfalfa	reduced lignin content	[229,230]

4.5. Genes Providing Resistance to Adverse Environmental Conditions

Rather than introducing a single stress resistance gene, scientists often utilize regulatory genes that function as "master switches" for plant stress tolerance. Such genes encode proteins that do not directly produce protective compounds but instead activate entire cascades of genes responsible for responding to drought, salinity, and extreme temperatures, both high and low. Examples of these genes include *DREB1* [231] and *HSP* [232], which trigger comprehensive defense systems already present within the plant. This approach is highly effective against a range of combined stresses. Other stress response genes include *ABI1*, *HAB1*, and *GSTU17*, which regulate the abscisic acid signaling pathway, thereby enhancing tolerance to drought and salinity [233]; the lipocalin *TIL* and *CHL* genes, which are essential for resistance to abiotic stress and for survival [234]; dehydrins (DHNs), which are activated in vegetative tissues of the plant and confer increased tolerance to drought and salt stress [235]; *eIF2a*, which is crucial for plant survival under conditions of macronutrient starvation [236]; *GATA16*, which improves cold tolerance at the seedling stage in rice [237] and others (see Table 6).

Table 6. Genes providing abiotic stress resistance.

GM Plant	Gene	Source	Desirable Trait	Ref.
rice, *Osteospermum ecklonis*, *Malus pumila* Mill., *Arabidopsis*	*myb4*	rice	resistance to cold, drought, and even certain diseases, changes in metabolite accumulation	[238–240]
Arabidopsis, tomato, wheat	*Zat12*	*Arabidopsis*, *Brassica carinata*	resistance to drought and low temperatures	[241–243]
tomato, *Arabidopsis*	*cold responsive-element binding factor 3* (*CBF3*)	*Arabidopsis*, *Punica granatum*, *Cuphea hookeriana*	cold resistance	[244–246]
wheat, tobacco, *Arabidopsis*	*Dehydration-Responsive Element-Binding 3* (*DREB3*)	soybean	increased drought tolerance	[247,248]

5. Global Status of GM Plants in the World

Earlier estimates from 2019 reported approximately 190 million hectares of GM crops cultivated across 29 countries, with soybean, maize, cotton, and canola being the predominant crops [5,249]. The adoption landscape had expanded by 2024: from 1996 to 2024, 73 countries had integrated GM crops through either cultivation or imports (Figure 4). In 2024 alone, more than 20 biotech/GM crops were cultivated across 31 countries, according to the ISAAA GM Approval Database (https://www.isaaa.org/gmapprovaldatabase/default.asp (accessed on 28 May 2026)).

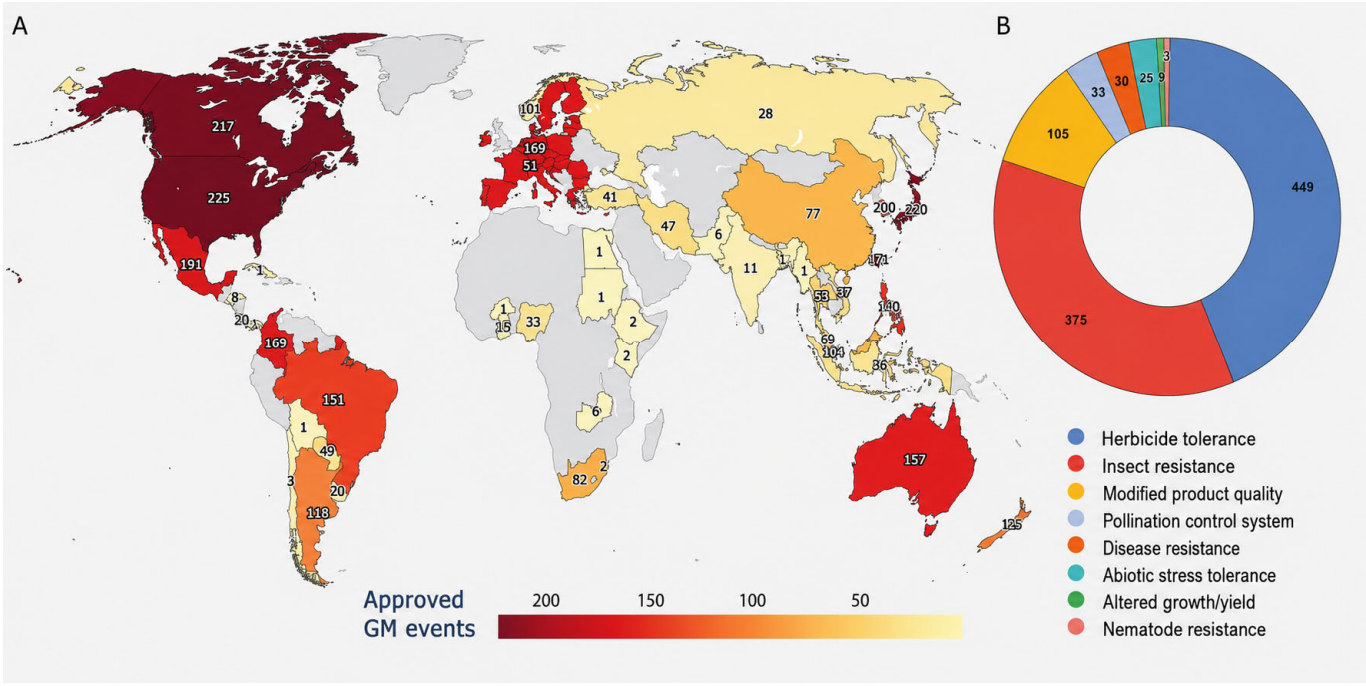


Figure 4. Distribution of approved GM crops. (**A**) Number of approved GM crop events by country or region according to the ISAAA GM Approval Database, accessed in May 2026. "Approved" indicates regulatory authorization for at least one use, such as cultivation, import, food/feed use, or commercialization, while a "GM event" refers to a specific GM plant line generated by a particular transformation event. Values should not be interpreted as the number of cultivars, plant species, or hectares under cultivation. (**B**) Distribution of approved GM crop events by commercial trait category according to the ISAAA GM Approval Database, accessed in May 2026. Event counts represent regulatory approval entries for specific GM events. Values should not be interpreted as numbers of commercial cultivars or hectares under cultivation. As stacked events may contain multiple traits, categories are not mutually exclusive.

6. Impact of GM Crops on Human Health

Each country has its own risk assessment protocol, which must be executed before a GM crop can be marketed globally. These protocols are designed to minimize risks to human health and prevent the displacement of natural crop varieties [250,251]. Different countries and authorities may employ their own methods to assess risks. These methods typically consider several factors: (1) how genetic modification can alter a plant's natural compounds or create entirely new ones; (2) whether the new compounds produced by the plant are safe; (3) analyzing the plant's metabolites; (4) examining how nutrients in the GM plant have changed. The risk assessment of GM crops concerning human health involves evaluating both direct effects (the GM plants themselves) and indirect effects (GM plants cultivated with chemicals).

6.1. The Direct Risk

A comprehensive analysis of health-focused studies reveals consistent safety profiles for approved GM crops across multiple evaluation parameters, including acute toxicity, subchronic effects, and nutritional equivalence [250,252]. Direct adverse effects on human health from the consumption of GM crops are now generally regarded as negligible [249]. Numerous studies have been conducted to assess the safety of GM rice, sugarcane, soybean, maize, and papaya in relation to human health. Scientists have investigated the impact of GM plants on health by feeding model animals (primarily rats and mice) with different GM crops over extended periods (ranging from 26 to 90 days, and in rare instances, up to 8 months). The feeding of model animals with GM plants, such as rice [253,254], sugarcane [255], maize [256–260], papaya [261], and soybean [262,263], has shown no mortality and only negligible changes in other biological parameters such as body weight, plasma protein levels, food utilization rate, and others. All GM plants were considered to be as safe as their non-GM counterparts [251]. Even in cases where transgenic soybeans contained three different resistance genes—conferring resistance to glyphosate (*EPSPS*), glufosinate (*pat*), and lepidopteran pests (*cry1Ac*, *cry2Ab2*, and *mVip3Aa*)—no effects were detected [264]. Studies on larger animals, such as pigs and calves fed with Bt maize, showed that Cry proteins were fragmented and diminished in the gastrointestinal tract and were not absorbed into the spleen, liver, or lymph nodes [265]. Moreover, heating maize prior to consumption further reduces exposure, and Cry proteins have not been detected in processed foods [266], confirming safety for human consumption [251]. A comprehensive seven-year feeding study on two generations of cynomolgus macaques found that GM maize with *Cry1Ab*/*Cry2Aj* and *EPSPS* genes had no major effects on gut microbiota composition, structure, or function [266]. This study is the longest GM crop feeding trial in non-human primates, offering key insights into multigenerational effects. Multigenerational reproductive toxicity studies have confirmed safety over long-term exposure. A three-generation rat study of GM maize with *Cry1Ab* and *EPSPS* genes also found no adverse effects on reproduction, offspring development, or multigenerational health [267]. Similarly, two-generation reproductive toxicity studies of *DREB3* GM wheat in rats demonstrated no treatment-related effects on reproductive function, fertility, or offspring viability [268,269].

6.2. The Indirect Risk

GM technology is based on resistance to pesticides, allowing farmers to spray them directly on GM crops. Pairing glyphosate with glyphosate-resistant GM crops removed a natural limitation on glyphosate use—glyphosate kills non-GM crops and targeted weeds. Relaxing this constraint has resulted in dramatic increases in glyphosate application intensity [270]. Glyphosate can affect human health through contaminated water, dust blown by the wind, aerial drift, and direct contact by consuming GM plants treated with glyphosate. Several independent studies have demonstrated that residues of glyphosate herbicides and its breakdown product, AMPA, accumulate in glyphosate-resistant plants [271,272]. The seeds of soybeans sprayed with the recommended glyphosate levels for weed control appear to have the highest reported levels of glyphosate of any food sourced from a glyphosate-resistant crop [273]. The impact of glyphosate on human health has been a matter of concern. While few scientists believe that the association of the herbicide with non-Hodgkin's lymphoma in humans is conclusive, others argue that current safety standards are outdated and need revision to prevent any long-term health effects from crops [274].

Detecting trace levels of glyphosate in human urine confirms systemic exposure [275]. The rise in endocrine and neurological disorders, such as Alzheimer's, has also been linked

to the increasing use of this pesticide in the USA [251,274]. The negative health impacts of glyphosate exposure in Brazil were demonstrated by Dias et al. [276] and Skidmore et al. [277]. They found that glyphosate exposure—driven by the expansion of GM seeds and transported through rivers—led to increased infant mortality and pediatric cancer deaths in Brazil. In the USA, it was shown that the introduction of GM seeds and glyphosate significantly reduced average birth weight and gestational length [270]. Bt crops provide resistance to lepidopteran insect pests, thereby reducing the need for chemical insecticide sprays against these specific pests. Numerous studies have indeed found evidence that the adoption of Bt crops is associated with significant reductions in insecticide use [278–280]. Furthermore, Bt maize contains a much lower concentration of highly hazardous mycotoxins than regular maize, making it considerably safer for human consumption [281]. Bt crops also offer direct health benefits to farmers through reduced insecticide exposure during spraying operations [281].

7. Discussion

The adoption of GM crops has been rapid and widespread across several major crop-producing countries around the world. Despite a vast number of GM plants known to possess trait genes for improved consumer qualities, pest and disease resistance, and enhanced productivity and immunity (Tables 2–5), adoption is primarily limited to three GM traits: herbicide tolerance, Bt tolerance, and product quality (Figure 4B), along with a small number of other commercial crops (from [249]). The use of GM plants in agriculture represents one of the important and contentious topics for discussion, as this approach has both significant advantages and serious drawbacks, as well as potential risks.

The advantages include increased yield attributable to specific transgenes. For instance, the presence of the Bt transgene in insect-resistant GM crops enables them to produce their own insecticidal proteins, rendering the plants toxic to insect pests. This reduces the need for chemical insecticides and leads to higher yields [282]. Resistance to viral diseases and adverse conditions, such as drought, also contributes to enhanced productivity. Another benefit of employing transgenic plants is the improvement of consumer qualities in agricultural crops. This might include an extended shelf life for fruit in transgenic tomatoes with delayed ripening or enhancements to nutritional value. One of the most renowned examples is Golden Rice, which is enriched with beta-carotene, a precursor of vitamin A (Table 5). Moreover, the improvement of nutritional value may encompass alterations in the composition of fatty acids, as demonstrated in transgenic soybeans. Additionally, by reducing the amount of machinery required to cultivate fields, carbon dioxide emissions into the atmosphere are decreased. The resilience of cultivated crops to adverse conditions (such as drought and soil salinity) also aids in preventing the expansion of agricultural land, thereby contributing to the preservation of forests. From an economic perspective, GM plants in agriculture offer further advantages. With reduced costs for chemical protection, fuel, and labor, this can result in decreased production costs. Furthermore, the use of GM plants tends to yield more predictable and stable results, as crop yields become less reliant on pests and unfavorable conditions.

However, the use of GM plants also carries certain risks. Firstly, there may be ecological risks. The introduction of glyphosate-resistant GM plants has led to a significant increase in the use of glyphosate, from 0.1 kg per hectare of cropland to over 1.3 kg per hectare in the USA [270]. Meanwhile, in the EU, which has never approved GM seeds, the rate of glyphosate application remains close to the United States' pre-GM levels, at approximately 0.2 kg per hectare [270]. The situation with herbicides mirrors the current scenario with antibiotics. Uncontrolled application of herbicides ultimately results in the evolution of herbicide-resistant weeds, specifically the emergence of so-called "superweeds". Conse-

quently, fields are treated with more toxic and dangerous herbicide mixtures, which leads to greater accumulation of herbicides and their breakdown products in GM plants, soil, water, and air.

Additionally, there are concerns that pollen from Bt plants may negatively affect beneficial insects, such as bees, or the soil microbiota. Furthermore, genes from transgenic crops may be transferred to wild relatives or organic crops, as controlling the spread of pollen by wind or insects is nearly impossible.

The application of GM plants also carries potential economic risks. Most often, the seeds of transgenic plants are patented by large corporations, which means that farmers must purchase seeds anew each season. This not only makes farmers dependent on multinational corporations, but also potentially leads to an increase in the so-called social divide, as smallholdings in developing countries often cannot afford expensive seeds and the accompanying herbicides.

There are concerns within society regarding the health risks associated with the consumption of GM plants, particularly concerning the potential allergenicity of such crops if the plant carries a gene for an allergen [283]. For example, a Brazil nut gene added to soybeans to boost nutrition triggered allergic reactions in tests [284].

Discussions also focus on the long-term effects of GM crops. The expansion of monocultures and the proliferation of resistant weeds and pests may diminish or even negate the short-term benefits of reduced insecticide use [285]. The long-term impact of GM crop adoption on species groups, including bees, butterflies, and other insects, remains largely unassessed. These groups may be directly affected by changes in GM crops and pesticide usage, and their abundance and diversity may, in turn, directly influence agricultural production [286]. Additionally, the quantification of the impact of GM crop adoption on global deforestation has yet to be undertaken [249]. Skepticism regarding GM crops persists, even in the absence of scientific evidence demonstrating harm from these products. Consequently, the current global approach is divided. In North and South America (the USA, Canada, Brazil, and Argentina), GM crops (such as soya, maize, rapeseed, and cotton) occupy extensive areas and form the backbone of export agriculture (Figure 4A). Conversely, in the EU, Russia, and various countries in Africa and Asia, stringent regulations are enforced concerning the cultivation of GM plants, with only certain crops permitted for import and processing (primarily for animal feed) or a moratorium in place, largely due to perceived environmental and health concerns.

8. Conclusions

Thus, GM plants, on the one hand, provide solutions to many problems related to hunger, deficiencies of specific nutrients in certain regions of the world, and the ability to grow and produce yields under unfavorable environmental conditions. On the other hand, the use of GM plants in agriculture carries certain economic and ecological risks. One of the challenges facing the global community at present is to balance the high potential of GM plants to improve agricultural efficiency with the management of the associated risks.

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Abbreviations

The following abbreviations are used in this manuscript:

GM	genetically modified
T-DNA	transfer DNA
Ti	tumour-inducing
(+)ssRNA	positive-strand
(−)ssRNA	negative-strand
VIGS	virus-induced gene silencing
VIGE	virus-induced genome editing
TSR	target-site resistance
NTSR	non-target-site resistance
AMPs	antimicrobial peptides
Bt	*Bacillus thuringiensis*
ISP	inhibitor of serine protease

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