

# Biotechnological Approaches for Enhancing Disease Resistance and Crop Quality in Horticultural Plants

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## Author's contribution

*This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.*

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## ABSTRACT

Horticulture plays a vital role in global food security, human nutrition, and economic development. However, horticultural crops face significant challenges from pests, diseases, and environmental stresses, leading to substantial yield losses. Conventional breeding methods have limitations in developing disease-resistant and high-yielding cultivars due to the narrow genetic base of horticultural crops and the time-consuming nature of traditional breeding. Biotechnological tools offer promising solutions to overcome these challenges and enhance crop productivity and disease resistance in horticulture. This review article explores the various biotechnological approaches, including marker-assisted selection (MAS), genetic engineering, genome editing, and micropropagation, and their applications in improving disease resistance and crop productivity in horticultural crops. MAS enables the precise and rapid selection of desired traits, such as disease resistance, by using molecular markers linked to the traits of interest. Genetic engineering allows the introduction of novel genes from diverse sources into horticultural crops to confer resistance against specific pathogens and pests. Genome editing technologies, particularly CRISPR/Cas9, provide a powerful tool for precise and targeted modifications of plant genomes to enhance disease resistance and other desirable traits. Micropropagation techniques facilitate the rapid multiplication of disease-free planting materials and the conservation of valuable germplasm. The article also discusses the challenges and future prospects of applying biotechnological tools in horticultural crop improvement. The integration of biotechnological approaches with conventional breeding and sustainable crop management practices holds great promise for developing disease-resistant and high-yielding horticultural crops, ensuring food security, and promoting sustainable horticulture in the face of global challenges.

*Keywords: Biotechnology; disease resistance; crop productivity; horticulture; molecular breeding.*

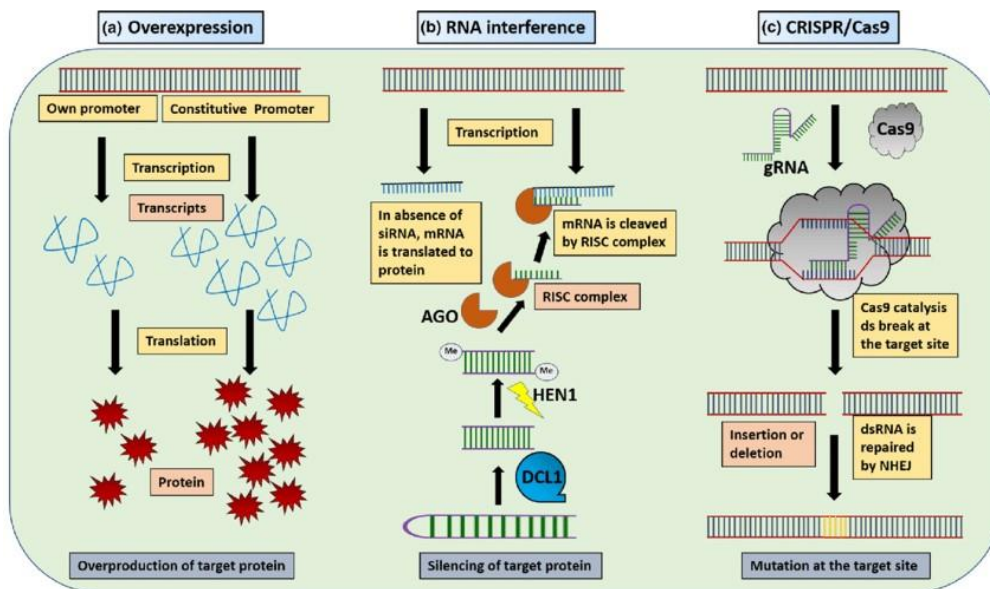
## 1. INTRODUCTION

Horticulture is a vital sector of agriculture that deals with the cultivation of fruits, vegetables, ornamental plants, and medicinal crops. It plays a crucial role in ensuring food and nutritional security, generating income, and promoting sustainable development worldwide [1]. However, horticultural crops are vulnerable to various biotic and abiotic stresses, including pests, diseases, and environmental factors, which can significantly reduce crop yield and quality [2]. Conventional breeding methods have been traditionally used to develop improved cultivars with enhanced disease resistance and productivity. However, these methods are time-consuming, labor-intensive, and limited by the available genetic diversity within the cultivated gene pool [3].

Biotechnological tools have emerged as powerful and innovative approaches to address the challenges faced by horticultural crops and to

accelerate the development of improved cultivars. These tools encompass a wide range of techniques, including marker-assisted selection (MAS), genetic engineering, genome editing, and micropropagation [4]. Biotechnology offers several advantages over conventional breeding methods, such as precise and targeted genetic manipulation, rapid generation of improved cultivars, and the ability to introduce novel traits from diverse genetic sources [5].

This review article aims to provide a comprehensive overview of the application of biotechnological tools in boosting disease resistance and crop productivity in horticulture. It will discuss the current status, advances, and future prospects of various biotechnological approaches and their potential to revolutionize the horticultural industry. The article will also highlight the challenges and ethical considerations associated with the deployment of biotechnology in horticulture.



**Fig. 1. Overview of biotechnological tools and their applications in horticultural crop improvement**

## 2. MARKER-ASSISTED SELECTION (MAS) FOR DISEASE RESISTANCE AND CROP IMPROVEMENT

Marker-assisted selection (MAS) is a powerful tool that integrates molecular markers with conventional breeding to accelerate the development of improved cultivars with desired traits, such as disease resistance and enhanced productivity [6]. Molecular markers are DNA sequences that are linked to specific genes or quantitative trait loci (QTLs) controlling the traits of interest. MAS allows breeders to select plants carrying the desired genes or QTLs based on the presence of linked molecular markers, without the need for extensive phenotypic evaluation [7].

### 2.1 Advantages of MAS in Horticultural Crop Improvement

MAS offers several advantages over conventional breeding methods in horticultural crop improvement:

1. Precision and efficiency: MAS enables the precise and early selection of plants carrying the desired genes or QTLs, reducing the time and resources required for phenotypic evaluation [8].
2. Acceleration of breeding cycles: MAS allows the selection of desired traits at the seedling stage, enabling the rapid generation of improved cultivars and reducing the breeding cycle time [9].

3. Pyramiding of multiple resistance genes: MAS facilitates the pyramiding of multiple disease resistance genes into a single cultivar, providing durable and broad-spectrum resistance against various pathogens [10].

Overcoming linkage drag: MAS helps in breaking the linkage between the desired trait and undesirable traits, enabling the development of superior cultivars with improved disease resistance and agronomic performance [11].

### 2.2 Application of MAS in Horticultural Crops

MAS has been successfully applied in various horticultural crops to improve disease resistance and crop productivity. Some notable examples include:

1. Tomato: MAS has been used to develop tomato cultivars resistant to various diseases, such as fusarium wilt, verticillium wilt, and tomato yellow leaf curl virus (TYLCV) [12]. For instance, the *Sw-5* gene, which confers resistance to tomato spotted wilt virus (TSWV), has been introgressed into elite tomato cultivars using MAS [13].
2. Potato: MAS has been employed to develop potato cultivars resistant to late blight, caused by the oomycete pathogen *Phytophthora infestans*. The *RB* gene,

derived from the wild potato species *Solanum bulbocastanum*, has been successfully introgressed into commercial potato cultivars using MAS, providing durable resistance against late blight [14].

3. Cucumber: MAS has been applied to develop cucumber cultivars resistant to downy mildew, caused by the fungal pathogen *Pseudoperonospora cubensis*. The *dm-1* gene, conferring resistance to downy mildew, has been incorporated into elite cucumber lines using MAS [15].
4. Citrus: MAS has been used to develop citrus cultivars resistant to citrus tristeza virus (CTV), a devastating viral disease. The *Ctv* resistance gene, derived from the trifoliolate orange (*Poncirus trifoliata*), has been introgressed into commercial citrus cultivars using MAS [16].

### 2.3 Challenges and Future Prospects of MAS in Horticulture

Despite the successful application of MAS in horticultural crop improvement, several challenges need to be addressed for its wider adoption:

1. **Marker-trait associations:** The effectiveness of MAS relies on the availability of robust and reliable marker-trait associations. Establishing these associations requires extensive genetic mapping and validation studies, which can be time-consuming and resource-intensive [20].
2. **Genetic background effects:** The expression of the desired trait may be influenced by the genetic background of the recipient cultivar. Therefore, the introgression of resistance genes through MAS may not always result in the expected level of resistance in different genetic backgrounds [21].
3. **Durability of resistance:** The durability of disease resistance conferred by single genes introgressed through MAS may be limited due to the evolution of new pathogen strains. Pyramiding multiple resistance genes and combining MAS with other disease management strategies can enhance the durability of resistance [22].

Future prospects of MAS in horticulture include the integration of advanced genomic tools, such as high-throughput sequencing and genotyping platforms, to accelerate marker development and trait mapping. The combination of MAS with

other biotechnological approaches, such as genome editing and genetic engineering, can further enhance the efficiency and precision of horticultural crop improvement [23].

### 3. GENETIC ENGINEERING FOR DISEASE RESISTANCE AND CROP PRODUCTIVITY

Genetic engineering involves the direct manipulation of an organism's genetic material by introducing foreign genes or modifying existing genes to confer desired traits [24]. In horticultural crops, genetic engineering has been widely used to develop transgenic plants with enhanced disease resistance, improved yield, and other desirable traits [25].

#### 3.1 Strategies for Genetic Engineering of Disease Resistance

Several strategies have been employed to genetically engineer disease resistance in horticultural crops:

1. **Pathogen-derived resistance (PDR):** PDR involves the introduction of genes derived from the pathogen itself into the plant genome. These genes, such as viral coat protein genes or viral replicase genes, interfere with the pathogen's replication or movement, conferring resistance to the plant [26].
2. **Plant defense genes:** The introduction of plant defense genes, such as those encoding pathogenesis-related (PR) proteins, chitinases, or glucanases, can enhance the plant's innate immune response against pathogens [27].
3. **Antimicrobial peptides (AMPs):** AMPs are small, cationic peptides with broad-spectrum antimicrobial activity. Transgenic plants expressing AMPs have shown enhanced resistance against various bacterial and fungal pathogens [28].
4. **RNA interference (RNAi):** RNAi is a gene silencing mechanism that can be exploited to develop transgenic plants resistant to viruses. Introducing virus-derived double-stranded RNA (dsRNA) into the plant triggers the degradation of the viral RNA, leading to resistance [29].

#### 3.2 Genetic Engineering for Crop Productivity

Genetic engineering has also been employed to enhance crop productivity in horticultural crops by targeting various traits:

1. Yield improvement: Transgenic approaches have been used to manipulate genes involved in photosynthesis, carbohydrate metabolism, and nutrient uptake to increase crop yield [30]. For example, the introduction of the *Arabidopsis* vacuolar H<sup>+</sup>-pyrophosphatase (*AVP1*) gene into tomato resulted in increased fruit yield under both normal and stress conditions [31].
2. Abiotic stress tolerance: Genetic engineering has been used to develop horticultural crops tolerant to abiotic stresses, such as drought, salinity, and extreme temperatures. The introduction of genes encoding stress-responsive transcription factors, osmolytes, or antioxidants has been shown to enhance stress tolerance in various horticultural crops [32].
3. Fruit quality improvement: Genetic engineering has been applied to improve fruit quality traits, such as shelf life, nutritional content, and sensory attributes. For instance, the suppression of the ripening-related gene *polygalacturonase* (*PG*) in tomato using antisense RNA technology resulted in delayed fruit softening and extended shelf life [33].

globally, with concerns about food safety, environmental impact, and ethical considerations. Addressing these concerns through transparent communication and rigorous safety assessments is crucial for the wider acceptance of GM horticultural crops [37].

2. Regulatory hurdles: The development and commercialization of GM crops are subject to strict regulatory frameworks, which can be time-consuming and costly. Harmonizing regulatory policies across countries and streamlining the approval process can facilitate the adoption of GM horticultural crops [38].
3. Intellectual property rights: The deployment of GM crops often involves complex intellectual property rights issues, with multiple patents covering different components of the technology. Navigating these issues and ensuring fair access to the technology for researchers and farmers is essential for the successful implementation of genetic engineering in horticulture [39].

Future prospects of genetic engineering in horticulture include the integration of advanced techniques, such as genome editing, to precisely modify plant genomes without introducing foreign genes. The combination of genetic engineering with other biotechnological tools, such as MAS and genomic selection, can accelerate the development of improved horticultural cultivars with enhanced disease resistance and crop productivity [40].

### 3.3 Challenges and Future Prospects of Genetic Engineering in Horticulture

Genetic engineering has the potential to revolutionize horticultural crop improvement, but it also faces several challenges:

1. Public acceptance: The public perception of genetically modified (GM) crops varies

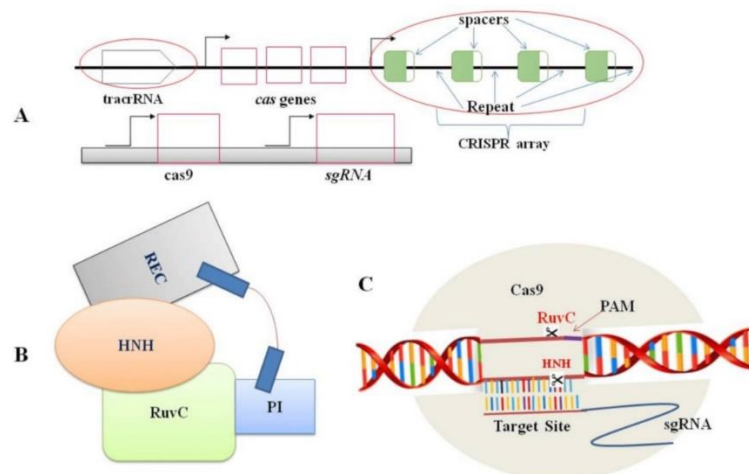


Fig. 2. Schematic representation of the CRISPR/Cas9 system for genome editing in plants

**Table 1. Examples of MAS application in horticultural crops for disease resistance**

Crop	Disease	Resistance Gene	Reference
Tomato	Fusarium wilt	<i>I-2</i>	[17]
Tomato	Verticillium wilt	<i>Ve</i>	[18]
Tomato	Tomato yellow leaf curl virus	<i>Ty-1, Ty-2, Ty-3</i>	[19]
Potato	Late blight	<i>RB</i>	[14]
Cucumber	Downy mildew	<i>dm-1</i>	[15]
Citrus	Citrus tristeza virus	<i>Ctv</i>	[16]

**Table 2. Examples of genetically engineered horticultural crops for disease resistance and crop productivity**

Crop	Trait	Transgene	Reference
Papaya	Papaya ringspot virus resistance	Viral coat protein gene	[34]
Banana	Fusarium wilt resistance	Chitinase gene ( <i>RCC2</i> )	[35]
Tomato	Increased fruit yield	<i>Arabidopsis</i> H <sup>+</sup> -pyrophosphatase ( <i>AVP1</i> )	[31]
Potato	Improved drought tolerance	Dehydration-responsive element binding protein ( <i>DREB1A</i> )	[36]
Tomato	Extended fruit shelf life	Antisense <i>polygalacturonase</i> ( <i>PG</i> )	[33]

**Table 3. Examples of CRISPR/Cas9 applications in horticultural crops**

Crop	Target Gene	Trait	Reference
Tomato	<i>MLO</i>	Powdery mildew resistance	[45]
Tomato	<i>SELF-PRUNING (SP)</i>	Compact plant architecture, increased yield	[46]
Tomato	<i>SISGR1</i>	Increased lycopene content	[47]
Cucumber	<i>eIF4E</i>	Potyvirus resistance	[48]
Banana	<i>PDS</i>	Albinism (proof-of-concept)	[49]

**Table 4. Examples of micropropagation applications in horticultural crops**

Crop	Micropropagation Technique	Application	Reference
Banana	Shoot tip culture	Virus elimination	[67]
Strawberry	Nodal culture	Rapid multiplication	[68]
Citrus	Somatic embryogenesis	Germplasm conservation	[69]
Potato	Shoot tip culture	Production of disease-free seed tubers	[70]
Orchids	Shoot tip culture	Rapid multiplication and conservation	[71]

#### 4. GENOME EDITING FOR PRECISE CROP IMPROVEMENT

Genome editing is a revolutionary biotechnological tool that enables precise and targeted modifications of plant genomes without introducing foreign DNA [41]. The most widely used genome editing technology is the clustered regularly interspaced short palindromic repeats (CRISPR)/CRISPR-associated protein 9 (Cas9) system, which has been successfully applied in various horticultural crops for trait improvement [42].

#### 4.1 CRISPR/Cas9 System: Mechanism and Applications

The CRISPR/Cas9 system consists of two main components: a guide RNA (gRNA) that directs the Cas9 endonuclease to a specific target site in the genome, and the Cas9 protein itself, which cleaves the DNA at the target site [43]. The resulting double-strand break (DSB) can be repaired through either non-homologous end joining (NHEJ) or homology-directed repair (HDR), leading to various types of genetic

modifications, such as gene knockouts, insertions, or replacements [44].

The CRISPR/Cas9 system has been widely used in horticultural crops for various applications:

1. Disease resistance: CRISPR/Cas9 has been used to target susceptibility genes or pathogen virulence factors to enhance disease resistance in horticultural crops. For example, the knockout of the *MLO* gene in tomato using CRISPR/Cas9 resulted in resistance to powdery mildew [45].
2. Yield improvement: CRISPR/Cas9 has been employed to modify genes involved in yield-related traits, such as plant architecture, flower development, and fruit size. In tomato, the modification of the *SELF-PRUNING* (*SP*) gene using CRISPR/Cas9 led to a compact plant architecture and increased fruit yield [46].
3. Nutritional enhancement: CRISPR/Cas9 has been used to improve the nutritional quality of horticultural crops by targeting genes involved in the biosynthesis or accumulation of specific nutrients. For instance, the knockout of the *S/SGR1* gene in tomato using CRISPR/Cas9 resulted in increased lycopene content in the fruit [47].

#### 4.2 Advantages of Genome Editing over Genetic Engineering

Genome editing, particularly the CRISPR/Cas9 system, offers several advantages over traditional genetic engineering approaches:

1. Precision and efficiency: CRISPR/Cas9 enables precise and targeted modifications of the plant genome, reducing off-target effects and increasing the efficiency of the desired genetic changes [50].
2. Multiplex editing: CRISPR/Cas9 allows the simultaneous targeting of multiple genes or genomic regions, enabling the modification of complex traits controlled by multiple loci [51].
3. Transgene-free products: Genome editing can produce plants with the desired genetic modifications without the integration of foreign DNA, potentially simplifying the regulatory approval process and increasing public acceptance [52].
4. Versatility: CRISPR/Cas9 can be used for various types of genetic modifications,

including gene knockouts, insertions, and replacements, providing greater flexibility in trait improvement compared to traditional genetic engineering approaches [53].

#### 4.3 Challenges and Future Prospects of Genome Editing in Horticulture

Despite the tremendous potential of genome editing in horticulture, several challenges need to be addressed:

1. Delivery methods: Efficient delivery of the CRISPR/Cas9 components into plant cells is crucial for successful genome editing. Developing optimized delivery methods, such as Agrobacterium-mediated transformation or particle bombardment, for various horticultural crops is essential [54].
2. Off-target effects: Although CRISPR/Cas9 is highly specific, off-target mutations can still occur. Minimizing off-target effects through careful gRNA design and using high-fidelity Cas9 variants is important to ensure the safety and effectiveness of the edited plants [55].
3. Regulatory uncertainty: The regulatory status of genome-edited crops varies among countries, with some regulating them as GMOs and others adopting more lenient approaches. Clarifying and harmonizing the regulatory frameworks for genome-edited crops will facilitate their development and commercialization [56].

Future prospects of genome editing in horticulture include the exploration of novel CRISPR systems, such as CRISPR/Cpf1 and CRISPR/Cas12a, which offer unique features and advantages compared to the CRISPR/Cas9 system [57]. The integration of genome editing with other biotechnological tools, such as genomics and bioinformatics, will further accelerate the precision and efficiency of horticultural crop improvement [58].

#### 5. MICROPROPAGATION FOR DISEASE-FREE PLANTING MATERIAL AND GERMPLASM CONSERVATION

Micropropagation is a biotechnological tool that involves the in vitro propagation of plants using small explants, such as shoot tips, nodal segments, or embryos, under sterile conditions [59]. Micropropagation offers several advantages

for horticultural crop production, including the rapid multiplication of disease-free planting material and the conservation of valuable germplasm [60].

### 5.1 Micropropagation Techniques and Applications

Various micropropagation techniques have been developed for horticultural crops, depending on the species and the desired outcome:

1. Shoot tip culture: Shoot tip culture involves the in vitro culture of apical meristems or shoot tips to produce multiple shoots, which can be rooted and acclimatized to produce complete plants [61].
2. Nodal culture: Nodal culture uses nodal segments containing axillary buds as explants for in vitro propagation. The axillary buds are induced to develop into shoots, which can be further multiplied and rooted [62].
3. Somatic embryogenesis: Somatic embryogenesis is the process of developing embryos from somatic cells in vitro. These embryos can be germinated to produce complete plants, providing a high multiplication rate and facilitating the production of synthetic seeds [63].

### 5.2 Micropropagation has Been Widely Applied in Horticultural Crops for Various Purposes

1. Disease elimination: Micropropagation, particularly shoot tip culture, can be used to produce virus-free planting material by exploiting the fact that many viruses do not invade the apical meristem [64].
2. Rapid multiplication: Micropropagation enables the rapid multiplication of elite genotypes or newly developed cultivars, allowing for the quick establishment of clonal plantations or the dissemination of improved planting material to farmers [65].
3. Germplasm conservation: Micropropagation can be used for the in vitro conservation of valuable germplasm, particularly for species with recalcitrant seeds or those that are difficult to conserve using conventional methods [66].

### 5.2 Challenges and Future Prospects of Micropropagation in Horticulture

Micropropagation has revolutionized the production of disease-free planting material and

the conservation of horticultural germplasm, but it also faces several challenges:

1. Genotype-dependent response: The success of micropropagation often depends on the genotype of the plant, with some genotypes being more responsive to in vitro culture than others. Optimizing the culture conditions for recalcitrant genotypes is necessary to expand the application of micropropagation [72].
2. Somaclonal variation: In vitro culture can sometimes lead to genetic or epigenetic changes in the propagated plants, known as somaclonal variation. Minimizing somaclonal variation through the use of appropriate culture conditions and the selection of stable genotypes is essential to maintain the genetic fidelity of the micropropagated plants [73].
3. Cost-effectiveness: The cost of micropropagation can be relatively high compared to conventional propagation methods, particularly for species with low multiplication rates or those requiring specialized culture conditions. Developing cost-effective micropropagation protocols and automating the process can help reduce the costs and increase the adoption of the technology [74].

Future prospects of micropropagation in horticulture include the integration of advanced biotechnological tools, such as molecular markers and cryopreservation, to enhance the efficiency and effectiveness of the technology [75]. The development of bioreactor systems for the large-scale production of micropropagated plants can further streamline the process and reduce the costs [76]. Additionally, the application of micropropagation for the production of secondary metabolites or the development of plant-based vaccines offers exciting opportunities for the pharmaceutical and biotechnology industries [77].

## 6. INTEGRATION OF BIOTECHNOLOGICAL TOOLS FOR COMPREHENSIVE CROP IMPROVEMENT

While each biotechnological tool discussed in this article has its own merits and applications, the integration of these tools can lead to a more comprehensive and effective approach to horticultural crop improvement. The synergistic



use of MAS, genetic engineering, genome editing, and micropropagation can accelerate the development of disease-resistant and high-yielding cultivars while ensuring the production of clean planting material and the conservation of valuable germplasm [78].

### **6.1 Combining MAS and Genetic Engineering**

The integration of MAS and genetic engineering can enhance the precision and efficiency of developing transgenic crops with improved disease resistance and agronomic traits. MAS can be used to identify and introgress natural resistance genes into elite cultivars, while genetic engineering can introduce novel resistance genes from diverse sources [79]. This combinatorial approach has been successfully applied in various horticultural crops, such as tomato, potato, and citrus, to develop cultivars with enhanced resistance to multiple pathogens [80].

### **6.2 Integrating Genome Editing and Micropropagation**

Genome editing and micropropagation can be integrated to develop improved cultivars and rapidly propagate them for commercial production. Genome editing can be used to precisely modify genes controlling disease resistance, yield, or quality traits, while micropropagation can facilitate the rapid multiplication and dissemination of the edited plants [81]. This approach has been demonstrated in various horticultural crops, such as banana, where CRISPR/Cas9-mediated genome editing was combined with micropropagation to develop and multiply disease-resistant lines [82].

### **6.3 Combining Multiple Biotechnological Tools**

The integration of multiple biotechnological tools, such as MAS, genetic engineering, genome editing, and micropropagation, can provide a comprehensive strategy for horticultural crop improvement. For example, MAS can be used to identify and introgress natural resistance genes, genetic engineering can introduce novel resistance genes, genome editing can precisely modify the introgressed genes or create new alleles, and micropropagation can facilitate the rapid multiplication and dissemination of the improved cultivars [83]. This multi-pronged

approach has the potential to revolutionize the development of disease-resistant and high-yielding horticultural crops.

## **7. CHALLENGES AND CONSIDERATIONS IN APPLYING BIOTECHNOLOGICAL TOOLS**

Despite the immense potential of biotechnological tools in horticultural crop improvement, several challenges and considerations need to be addressed for their successful application and widespread adoption.

### **7.1 Technical Challenges**

1. **Genotype-dependent response:** The efficiency and success of biotechnological tools, such as genetic transformation and genome editing, often vary depending on the genotype of the crop species or cultivar [87]. Developing genotype-independent protocols or optimizing the techniques for specific genotypes is crucial for the broader application of these tools.
2. **Regeneration and transformation bottlenecks:** Many horticultural crops, particularly woody perennial species, are recalcitrant to *in vitro* regeneration and genetic transformation [88]. Overcoming these bottlenecks through the optimization of regeneration protocols and the development of efficient transformation methods is essential for the successful application of biotechnological tools.
3. **Off-target effects and unintended consequences:** Genetic engineering and genome editing can sometimes lead to off-target effects or unintended consequences, such as pleiotropic effects on non-target traits [89]. Careful design of constructs, thorough screening of transgenic or edited plants, and comprehensive risk assessment are necessary to minimize these undesired effects.

### **7.2 Regulatory and Socioeconomic Considerations**

1. **Regulatory frameworks:** The regulatory approval process for genetically modified or edited crops varies among countries, with some having more stringent regulations than others [90]. Harmonizing the regulatory frameworks and establishing science-based, transparent, and

predictable approval processes can facilitate the commercialization and adoption of improved cultivars developed through biotechnological tools.

2. Public acceptance: Public perception and acceptance of genetically modified or edited crops can greatly influence their adoption and marketability [91]. Effective science communication, stakeholder engagement, and transparent sharing of information about the benefits, risks, and safety of these crops are essential to build public trust and support for biotechnology-derived products.
3. Intellectual property rights and access: The development and application of biotechnological tools often involve intellectual property rights, such as patents, which can limit access to these technologies for researchers and farmers, particularly in developing countries [92]. Establishing mechanisms for fair and equitable sharing of benefits, promoting open innovation, and creating public-private partnerships can help ensure broader access to these tools and technologies.

### 7.3 Integrated Approach and Sustainable Deployment

1. Integration with conventional breeding: Biotechnological tools should be integrated with conventional breeding programs to harness the strengths of both approaches [93]. Molecular breeding strategies, such as marker-assisted backcrossing and genomic selection, can facilitate the introgression of desirable traits from biotechnology-derived lines into elite cultivars and breeding populations.
2. Sustainable crop management practices: The deployment of disease-resistant and high-yielding cultivars developed through biotechnological tools should be accompanied by sustainable crop management practices, such as integrated

pest management, soil health management, and efficient irrigation systems [94]. This holistic approach can ensure the long-term sustainability and resilience of horticultural production systems.

3. Capacity building and technology transfer: Strengthening the capacity of researchers, extension workers, and farmers in applying biotechnological tools and best practices is crucial for their successful adoption and impact [95]. Promoting technology transfer, regional collaborations, and knowledge sharing among stakeholders can accelerate the dissemination and uptake of these tools and technologies.

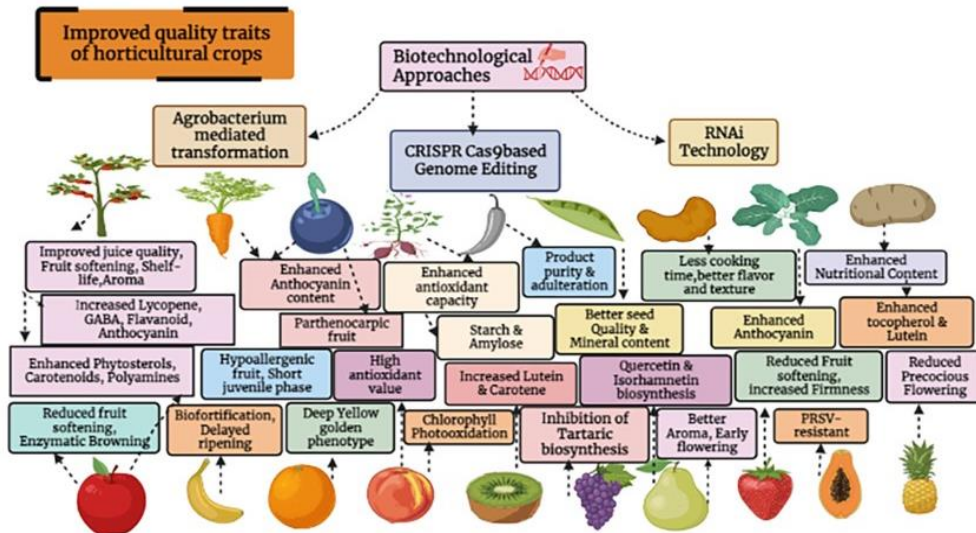
## 8. FUTURE OUTLOOK

Biotechnological tools, including marker-assisted selection, genetic engineering, genome editing, and micropropagation, hold immense potential for revolutionizing horticultural crop improvement. These tools offer unprecedented opportunities to develop disease-resistant and high-yielding cultivars, produce clean planting material, and conserve valuable germplasm. The integration of these tools can provide a comprehensive and effective strategy for addressing the complex challenges faced by horticultural production systems.

However, the successful application and widespread adoption of biotechnological tools in horticulture require addressing various technical, regulatory, and socioeconomic challenges. Continued research and development efforts are needed to optimize these tools for specific crops and genotypes, minimize off-target effects, and develop efficient regeneration and transformation protocols. Establishing science-based and harmonized regulatory frameworks, promoting public acceptance through effective communication and engagement, and ensuring fair access to these technologies are crucial for their responsible and sustainable deployment.

**Table 5. Examples of integrating biotechnological tools in horticultural crops**

Crop	Integrated Tools	Application	Reference
Tomato	MAS + Genetic Engineering	Multiple disease resistance	[84]
Potato	MAS + Genetic Engineering	Late blight and virus resistance	[85]
Banana	Genome Editing + Micropropagation	Fusarium wilt resistance	[82]
Citrus	MAS + Genetic Engineering + Micropropagation	Huanglongbing resistance	[86]



**Fig. 3. Integration of biotechnological tools for comprehensive horticultural crop improvement**

The future outlook for biotechnology in horticulture is promising, with rapid advancements in genomics, bioinformatics, and precision breeding techniques. The integration of these cutting-edge technologies with conventional breeding and sustainable crop management practices can accelerate the development of climate-resilient, nutrient-efficient, and disease-resistant horticultural crops. Furthermore, the application of biotechnology for the production of high-value compounds, such as pharmaceuticals, biofuels, and specialty chemicals, can open up new avenues for the horticultural industry.

## 9. CASE STUDIES OF SUCCESSFUL BIOTECHNOLOGICAL INTERVENTIONS IN HORTICULTURE

This section can provide specific examples of how biotechnological tools have been successfully applied to improve disease resistance and crop productivity in horticultural crops. These case studies can highlight the impact of these interventions on farmers, consumers, and the industry as a whole.

### 9.1 Bt Eggplant in Bangladesh

Bt eggplant, developed through genetic engineering to resist the destructive fruit and shoot borer pest, has been successfully adopted by farmers in Bangladesh. The adoption of Bt eggplant has led to significant reductions in pesticide use, increased yields, and improved farmer incomes [96]. This case study

demonstrates the potential of genetic engineering to address critical pest problems in horticultural crops and improve the livelihoods of smallholder farmers.

### 9.2 Virus-Resistant Papaya in Hawaii

The papaya industry in Hawaii was severely threatened by the papaya ringspot virus (PRSV) in the 1990s. Researchers developed a genetically engineered papaya variety, 'Rainbow,' resistant to PRSV using the pathogen-derived resistance approach [97]. The adoption of 'Rainbow' papaya saved the Hawaiian papaya industry and has been hailed as a success story of biotechnology in horticulture.

### 9.3 Marker-Assisted Selection for Fusarium Wilt Resistance in Tomato

Marker-assisted selection has been successfully used to develop tomato varieties resistant to Fusarium wilt, a devastating fungal disease. By using molecular markers linked to resistance genes, breeders have been able to efficiently introgress these genes into elite tomato cultivars, resulting in the development of resistant varieties that are widely grown by farmers [98]. This case study highlights the power of marker-assisted selection in accelerating the development of disease-resistant cultivars.

## 10. THE ROLE OF BIOTECHNOLOGY IN ADDRESSING GLOBAL CHALLENGES IN HORTICULTURE

Biotechnological tools have the potential to address various global challenges faced by the

horticultural sector, such as climate change, resource scarcity, and the need for more nutritious and sustainable food systems.

### **10.1 Climate Change Adaptation**

Climate change poses significant threats to horticultural production, including increased abiotic stresses, altered pest and disease pressures, and reduced water availability [99]. Biotechnological tools can be used to develop crop varieties that are more resilient to these stresses. For example, genetic engineering and genome editing can be used to introduce genes or modify existing genes to enhance drought tolerance, heat tolerance, and disease resistance in horticultural crops [100]. Marker-assisted selection can also be used to identify and introgress traits associated with climate resilience from wild relatives or landraces into elite cultivars.

### **10.2 Resource Use Efficiency**

Increasing resource use efficiency, particularly in terms of water and nutrients, is crucial for sustainable horticultural production. Biotechnological tools can be used to develop crop varieties with improved water and nutrient use efficiency. For instance, genetic engineering can be used to introduce genes that enhance root growth, water uptake, and nutrient acquisition [101]. Genome editing can be employed to modify genes involved in stomatal regulation, photosynthesis, and nutrient transport to optimize resource use efficiency [102]. Micropropagation techniques can also be used to produce planting materials with uniform and efficient root systems.

### **10.3 Biofortification for Nutritional Security**

Horticultural crops are important sources of essential nutrients, vitamins, and minerals for human health. However, many populations worldwide suffer from micronutrient deficiencies due to limited access to diverse and nutritious diets [103]. Biotechnological tools can be used to enhance the nutritional content of horticultural crops through biofortification. Genetic engineering and genome editing can be used to increase the levels of specific nutrients, such as vitamins, minerals, and essential amino acids, in crops [104]. For example, golden rice, a genetically engineered variety of rice enriched with beta-carotene, has been developed to

address vitamin A deficiency in developing countries [105].

## **11. ETHICAL AND SOCIETAL CONSIDERATIONS**

The application of biotechnological tools in horticulture raises various ethical and societal considerations that need to be addressed to ensure responsible and equitable deployment of these technologies.

### **11.1 Equitable Access and Benefit Sharing**

The development and commercialization of biotechnology-derived products often involve intellectual property rights and patents, which can limit access to these technologies for researchers, farmers, and consumers, particularly in developing countries [106]. Ensuring equitable access to the benefits of biotechnological innovations is crucial for promoting food security and reducing inequalities. Mechanisms such as humanitarian licensing, patent pooling, and public-private partnerships can be explored to facilitate the sharing of technologies and benefits [107].

### **11.2 Socioeconomic Impacts**

The adoption of biotechnology-derived crops can have significant socioeconomic impacts on farmers, communities, and the wider society. While these crops can potentially increase yields, reduce costs, and improve livelihoods, they may also lead to unintended consequences, such as the displacement of traditional varieties, the concentration of market power, and the marginalization of smallholder farmers [108]. Assessing and mitigating the potential socioeconomic impacts of biotechnological interventions through participatory and inclusive approaches is essential for ensuring their sustainable and equitable deployment.

### **11.3 Ethical Considerations**

The application of biotechnological tools in horticulture raises ethical questions related to the manipulation of living organisms, the safety and long-term impacts of genetically modified or edited crops, and the potential ecological risks associated with the release of these crops into the environment [109]. Addressing these ethical concerns requires transparent and inclusive

public dialogue, rigorous risk assessment and management, and the development of appropriate governance frameworks that balance the benefits and risks of these technologies [110].

### **13. CAPACITY BUILDING AND KNOWLEDGE TRANSFER FOR BIOTECHNOLOGY ADOPTION**

For the successful adoption and implementation of biotechnological tools in horticulture, it is crucial to build the capacity of researchers, extension workers, farmers, and other stakeholders. This section will discuss the importance of capacity building and knowledge transfer activities to promote the uptake of biotechnology in the horticultural sector.

#### **13.1 Training and Education Programs**

Providing training and education programs on biotechnological tools and their applications is essential for building the skills and knowledge of stakeholders involved in horticultural research and production. These programs can include workshops, seminars, and hands-on training sessions covering topics such as molecular marker techniques, genetic engineering, genome editing, and micropropagation [111]. Collaboration between research institutions, universities, and extension services can facilitate the development and delivery of these training programs.

#### **13.2 Extension Services and Farmer Outreach**

Extension services play a vital role in disseminating information and technologies to farmers and promoting their adoption. Strengthening the capacity of extension workers to effectively communicate the benefits and risks of biotechnological tools to farmers is crucial for their successful implementation [112]. Farmer outreach activities, such as field demonstrations, participatory research, and farmer field schools, can help engage farmers in the technology development and adoption process, ensuring that their needs and concerns are addressed [113].

#### **13.3 International Collaborations and Knowledge Sharing**

Fostering international collaborations and knowledge sharing among researchers, institutions, and countries can accelerate the

development and adoption of biotechnological tools in horticulture. Collaborative research projects, scientific exchanges, and networking platforms can facilitate the exchange of expertise, resources, and best practices [114]. Regional and global initiatives, such as the Consultative Group on International Agricultural Research (CGIAR) and the International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRFA), can support these efforts and promote the sharing of knowledge and genetic resources [115].

### **14. BIOTECHNOLOGY FOR SUSTAINABLE HORTICULTURE AND ECOSYSTEM SERVICES**

Biotechnological tools can contribute to the development of sustainable horticultural systems that provide ecosystem services and support biodiversity conservation. This section will explore the potential applications of biotechnology in promoting sustainable horticulture and enhancing ecosystem services.

#### **14.1 Biodiversity Conservation and Utilization**

Horticultural crops are an important component of agricultural biodiversity, and their conservation and sustainable utilization are crucial for food security and resilience. Biotechnological tools, such as molecular markers and cryopreservation, can support the characterization, conservation, and utilization of horticultural genetic resources [116]. For example, molecular markers can be used to assess the genetic diversity of crop wild relatives and landraces, informing strategies for their conservation and incorporation into breeding programs [117].

#### **14.2 Agroecological Approaches and Biotechnology**

Integrating biotechnological tools with agroecological approaches can contribute to the development of sustainable and resilient horticultural systems. Agroecological practices, such as intercropping, agroforestry, and conservation biological control, can be complemented by the use of biotechnology-derived crops that are resistant to pests and diseases, reducing the need for chemical inputs [118]. Additionally, biotechnological tools can be used to study and harness the beneficial

interactions between crops and their associated microbiomes, supporting the development of biofertilizers and biopesticides [119].

### **14.3 Ecosystem Services and Landscape-Level Benefits**

Horticultural crops can provide various ecosystem services, such as pollination, soil conservation, carbon sequestration, and habitat provision for beneficial organisms. Biotechnological tools can be used to enhance the ability of horticultural crops to deliver these services. For example, genetic engineering and genome editing can be used to develop crops with enhanced floral traits that attract pollinators [120] or with improved root systems that contribute to soil health and carbon storage [121]. At the landscape level, the deployment of biotechnology-derived crops with resistance to pests and diseases can reduce the spread of these threats to natural ecosystems, supporting biodiversity conservation [122].

## **15. PUBLIC-PRIVATE PARTNERSHIPS AND STAKEHOLDER ENGAGEMENT**

The successful development and deployment of biotechnological tools in horticulture require effective public-private partnerships and stakeholder engagement. This section will discuss the importance of these collaborations and strategies for fostering inclusive innovation systems.

### **15.1 Public-Private Partnerships for Research and Development**

Public-private partnerships (PPPs) are essential for leveraging the strengths and resources of both the public and private sectors in the research and development of biotechnological tools for horticulture. PPPs can facilitate the sharing of knowledge, expertise, and facilities, accelerating the development and commercialization of innovative technologies [123]. These partnerships can also help ensure that the developed technologies are accessible and affordable for smallholder farmers and resource-poor communities [124].

### **15.2 Stakeholder Engagement and Participatory Approaches**

Engaging stakeholders, including farmers, consumers, researchers, policymakers, and civil society organizations, in the development and

deployment of biotechnological tools is crucial for ensuring their relevance, acceptability, and adoption. Participatory approaches, such as participatory plant breeding and participatory technology development, can help align the development of biotechnological tools with the needs and preferences of end-users [125]. Stakeholder engagement can also facilitate the co-creation of knowledge, build trust, and promote the responsible and equitable use of these technologies [126].

### **15.3 Inclusive Innovation Systems**

Fostering inclusive innovation systems that involve and benefit all stakeholders, particularly smallholder farmers and marginalized communities, is essential for the equitable and sustainable adoption of biotechnological tools in horticulture. This requires creating an enabling environment that supports the participation of diverse stakeholders, promotes access to information and resources, and ensures the fair sharing of benefits [127]. Strategies such as capacity building, participatory research, and multi-stakeholder platforms can contribute to the development of inclusive innovation systems [128].

## **16. CONCLUSION**

The application of biotechnological tools in horticulture has the potential to revolutionize crop improvement, enhance food and nutritional security, and promote sustainable agricultural practices. Marker-assisted selection, genetic engineering, genome editing, and micropropagation have already demonstrated their ability to develop disease-resistant and high-yielding cultivars, produce clean planting material, and conserve valuable germplasm. The integration of these tools with other emerging technologies and agroecological approaches can further contribute to the development of resilient and sustainable horticultural systems. However, realizing the full potential of biotechnology in horticulture requires addressing various technical, regulatory, socioeconomic, and ethical challenges. Continued research and development efforts are needed to refine and optimize these tools for specific crops and contexts, assess and manage potential risks, and ensure their responsible and equitable deployment. Capacity building, knowledge transfer, and stakeholder engagement are crucial for fostering the adoption and uptake of these technologies by farmers and other stakeholders.

Public-private partnerships and inclusive innovation systems are essential for leveraging the strengths and resources of different actors and ensuring that the benefits of biotechnological tools are accessible and equitably shared. By working together across disciplines, sectors, and stakeholder groups, we can harness the potential of biotechnology to build a more sustainable, resilient, and inclusive horticultural future. As we move forward, it is important to recognize that biotechnology is not a silver bullet solution but rather a set of tools that must be used in conjunction with other approaches, such as agroecology, sustainable crop management, and socioeconomic interventions, to address the complex challenges faced by the horticultural sector. By taking a holistic and integrated approach, we can ensure that the application of biotechnology in horticulture supports the achievement of the Sustainable Development Goals and contributes to a more food-secure and sustainable world.

#### DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of manuscripts.

#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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