

Review Article

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Molecular Breeding for Pest and Disease Resistance in Legumes

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Abstract This study focuses on the application of molecular breeding techniques to enhance pest and disease resistance in legume plants, which are critical for global food security and agricultural sustainability, identifies and utilizes molecular markers associated with resistance traits, analyzes genetic diversity, and implements advanced breeding strategies such as marker-assisted selection (MAS) and genomic selection. By identifying key resistance genes and the successful application of MAS, significant progress has been made in developing resistant bean varieties that improve pest and disease resistance, the results highlight the potential of molecular breeding in revolutionizing bean production, ensuring higher yields and resilience to biological stresses.

Keywords Molecular breeding; Legumes; Pest resistance; Disease resistance; Marker-assisted selection

1 Introduction

Legumes are a cornerstone of global agriculture, playing a critical role in ensuring food security and nutritional health. They are a primary source of protein for both human and livestock consumption, particularly in developing countries where access to animal protein may be limited (Pandey et al., 2016; Varshney et al., 2018; Zhang, 2024). Additionally, legumes contribute significantly to soil health through their ability to fix atmospheric nitrogen, thereby reducing the need for synthetic fertilizers and promoting sustainable agricultural practices (Ramalingam et al., 2015; Bueno and Lopes, 2020). The cultivation of legumes such as chickpea, soybean, and common bean is essential not only for their nutritional benefits but also for their environmental advantages, including biodiversity preservation and soil fertility enhancement (Roriz et al., 2020; Wohor et al., 2022).

Despite their importance, legume production faces significant challenges from various pests and diseases, which can lead to substantial yield losses and threaten food security. Viral diseases, for instance, are a major biotic stress that severely impacts grain legume production globally (Jha et al., 2023). Similarly, soilborne pathogens such as those causing rhizospheric diseases in peas are difficult to manage and persist in the soil for long periods, complicating control efforts (Wohor et al., 2022). Ascochyta blight is another critical disease affecting grain legumes, causing considerable yield reductions worldwide (Jha et al., 2022). The increasing incidence of these diseases, exacerbated by climate change, underscores the urgent need for effective resistance breeding strategies (Pratap et al., 2021).

This study will explore the use of genomic resources, marker-assisted selection, and cutting-edge biotechnology tools such as CRISPR/Cas9 to achieve durable resistance in legume breeding programs, with the hope of supporting sustainable agriculture and ensuring food security by mitigating the impact of pests and diseases on legume crops. This study aims to explore molecular breeding methods for improving resistance to pests and diseases in legume plants, identify and deploy genetic determinants of resistance to various pests and diseases by leveraging advances in genomics, phenotyping, and bioinformatics, and integrate next-generation sequencing techniques, high-throughput genotyping, and functional genomics to accelerate the development of resistant legume varieties.

2 Overview of Pests and Diseases Affecting Legumes

2.1 Common pests affecting legume crops

Legume crops are susceptible to a variety of pests that can significantly impact their yield and quality. Among the most common pests are aphids and pod borers. Aphids are sap-sucking insects that can cause direct damage by feeding on plant sap and indirect damage by transmitting viral diseases. Pod borers, such as the legume pod borer (*Maruca vitrata*), are notorious for their destructive feeding habits on flowers, pods, and seeds, leading to substantial yield losses (Rubiales et al., 2015; Jha et al., 2023).

2.2 Major diseases impacting legumes

Legume crops are affected by a wide range of diseases caused by fungal, bacterial, and viral pathogens. Fungal diseases such as rusts, powdery and downy mildews, ascochyta blights, botrytis gray molds, anthracnoses, damping-off, root rots, collar rots, vascular wilts, and white molds are particularly devastating (Rubiales et al., 2015; Pandey et al., 2023). Bacterial diseases, including common bacterial blight, halo blight, and bacterial brown spot, also pose significant threats to legume production. Viral diseases, such as Bean common mosaic virus (BCMV), Bean golden mosaic virus (BGMV), and Bean golden yellow mosaic virus (BGYMV), further complicate the management of legume crops (Jha et al., 2023).

2.3 Economic and yield losses due to pest and disease outbreaks in legume crops

Pest and disease outbreaks in legume crops can lead to severe economic and yield losses. For instance, diseases like anthracnose and ascochyta blight can cause significant yield reductions, sometimes up to 100% in severe cases (Jha et al., 2022; Pandey et al., 2023). The economic impact is profound, as legumes are a crucial source of protein and income for many farmers, especially in developing countries. The cost of managing these pests and diseases, including the use of fungicides, insecticides, and resistant varieties, adds to the financial burden on farmers (Rubiales et al., 2015; Pandey et al., 2023). Additionally, the continuous evolution of pathogens and pests under high selection pressure necessitates ongoing research and development of new resistant cultivars to sustain legume production (Martins et al., 2020; Saxena et al., 2023).

3 Conventional Breeding for Pest and Disease Resistance

3.1 Traditional breeding strategies used to develop resistant legume varieties

Traditional breeding strategies for developing pest and disease-resistant legume varieties have primarily relied on the selection and cross-breeding of plants exhibiting desirable traits (Zhou and Guo, 2024). Phenotypic selection involves selecting plants that show natural resistance to pests and diseases and using them as parents in breeding programs. For example, in common beans, breeders have used phenotypic selection to develop cultivars with enhanced levels of disease and pest resistance (Basavaraja et al., 2020). Cross-breeding between resistant and susceptible varieties to combine desirable traits. In lentils, wide hybridization and ovule rescue techniques have been employed to introgress resistance traits from wild relatives into cultivated varieties. Induced mutagenesis creates genetic variability through mutagenesis to develop disease-resistant lines. This approach has been used in lentils to develop several disease-resistant mutant lines (Roy et al., 2023). Utilize related species to introduce resistance genes. For instance, interspecific hybridizations with *Phaseolus coccineus* and *Phaseolus acutifolius* have been used to introduce disease resistance in common beans.

3.2 Limitations of conventional breeding in achieving durable resistance

Despite the successes, conventional breeding faces several limitations in achieving durable resistance. The process of developing resistant varieties through traditional breeding methods can take 7~10 years, requiring significant economic resources (Basavaraja et al., 2020). Traits such as resistance to multiple pathogens and environmental stressors are often complex and controlled by multiple genes, making them difficult to select and breed for using conventional methods (Basavaraja et al., 2020). The effectiveness of resistance traits can vary significantly across different environments, complicating the selection process. The genetic base of cultivated varieties is often narrow, limiting the availability of resistance genes (Rubiales et al., 2015). Pathogens can evolve and overcome resistance, necessitating continuous breeding efforts to keep up with new strains (Wille et al., 2018).

3.3 Examples of successful disease-resistant legume varieties developed through traditional breeding

Several successful disease-resistant legume varieties have been developed through traditional breeding methods. Breeding programs have developed common bean varieties resistant to major diseases such as anthracnose, angular leaf spot, and common bacterial blight. These programs have utilized phenotypic selection and interspecific hybridization to achieve these results (Basavaraja et al., 2020). Disease-resistant lentil varieties have been developed using wide hybridization and induced mutagenesis. These varieties exhibit resistance to various diseases, contributing to improved productivity and resilience (Roy et al., 2023). Traditional breeding has led to the development of soybean varieties resistant to anthracnose, with quantitative trait loci (QTLs) identified for resistance, enabling marker-assisted selection (Figure 1) (Pandey et al., 2023). These examples highlight the potential of conventional breeding to develop disease-resistant legume varieties, although the integration of molecular tools can further enhance the efficiency and effectiveness of these efforts.

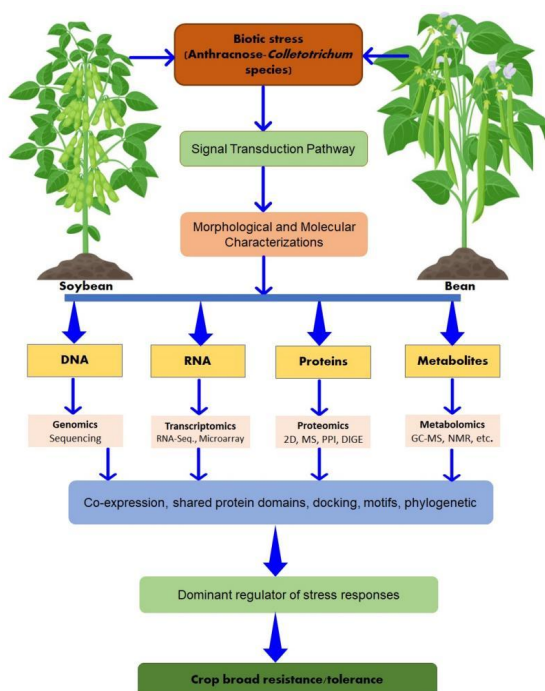


Figure 1 Schematic explanation of omics approaches used in future projects in the improvement of resistance/tolerant to legume anthracnose (PPI: protein-protein interaction; DIGE: differential gel electrophoresis; GC-MS: gas chromatography-mass spectrometry) (Adopted from Pandey et al., 2023)

4 Introduction to Molecular Breeding

4.1 Definition and key principles of molecular breeding

Molecular breeding refers to the application of molecular biology tools, particularly DNA markers, to assist in the selection and breeding of plants with desirable traits. This approach leverages advancements in genomics and biotechnology to enhance the efficiency and precision of traditional breeding methods. Key principles of molecular breeding include the identification and utilization of molecular markers linked to specific traits, the development of genetic maps, and the application of marker-assisted selection (MAS) to accelerate the breeding process (Varshney et al., 2013; Basavaraja et al., 2020).

4.2 Comparison between conventional and molecular breeding approaches

Conventional breeding relies on phenotypic selection, which can be time-consuming and less precise due to the influence of environmental factors on trait expression. It typically involves crossing plants with desirable traits and selecting the best offspring over multiple generations, a process that can take several years (Basavaraja et al., 2020). In contrast, molecular breeding uses DNA markers to identify and select plants with the desired genetic traits at an early stage, significantly reducing the breeding cycle time. This method allows for the precise introgression of traits such as disease resistance, drought tolerance, and improved yield (Varshney et al., 2013; Jha

et al., 2022). For example, marker-assisted backcrossing (MABC) has been successfully used to introgress resistance to *Fusarium* wilt and *Ascochyta* blight in chickpea, and late leaf spot and nematode resistance in groundnut (Varshney et al., 2013).

4.3 Role of molecular markers in pest and disease resistance breeding

Molecular markers play a crucial role in breeding for pest and disease resistance by enabling the identification of genomic regions associated with resistance traits. These markers can be used in various breeding strategies, including MAS, marker-assisted backcrossing (MABC), and genomic selection (GS) (Varshney et al., 2013; Jha et al., 2023). For instance, in common bean, molecular markers have been used to develop resistance to multiple diseases such as angular leaf spot, anthracnose, and common bacterial blight (Meziadi et al., 2016; Jha et al., 2022). The use of next-generation sequencing (NGS) technologies has further enhanced the development of high-density genetic maps and the identification of quantitative trait loci (QTLs) associated with disease resistance, facilitating the rapid development of resistant cultivars (Yang et al., 2012; Kankanala et al., 2019). Additionally, the integration of genomics and functional genomics approaches has helped uncover the molecular mechanisms underlying resistance, providing valuable insights for breeding programs (Kankanala et al., 2019; Jha et al., 2023).

5 Marker-Assisted Selection (MAS) for Disease and Pest Resistance

5.1 Overview of MAS in legume breeding programs

Marker-assisted selection (MAS) has revolutionized legume breeding programs by enabling the precise and efficient selection of desirable traits, particularly those related to disease and pest resistance. MAS leverages molecular markers linked to specific genes or quantitative trait loci (QTLs) to facilitate the identification and incorporation of resistance traits into new cultivars. This approach has been particularly beneficial in legumes, where traditional breeding methods are often hampered by the complex inheritance patterns of resistance traits and the influence of environmental factors on phenotypic expression (Varshney et al., 2013).

5.2 Identification and use of molecular markers linked to resistance genes

The identification of molecular markers linked to resistance genes is a critical step in MAS. Various types of markers, such as simple sequence repeats (SSRs), amplified fragment length polymorphisms (AFLPs), and restriction fragment length polymorphisms (RFLPs), have been employed to map resistance genes in legumes. For instance, in common bean, markers linked to resistance genes for diseases like angular leaf spot, anthracnose, and Bean common mosaic virus have been identified and utilized in breeding programs. Similarly, in lupin, markers linked to anthracnose resistance have been developed and validated for use in MAS. These markers enable breeders to select for resistance traits even in the absence of the pathogen, thereby accelerating the breeding process and improving the efficiency of resistance gene incorporation (Wu et al., 2022).

5.3 Success stories of MAS in improving pest and disease resistance in legume crops

Several success stories highlight the effectiveness of MAS in enhancing pest and disease resistance in legume crops. In mung bean, MAS has been used to introgress bruchid resistance genes, resulting in the development of new resistant cultivars with improved agronomic traits (Wu et al., 2022). In chickpea, MAS has facilitated the incorporation of resistance to *Fusarium* wilt and *Ascochyta* blight, significantly improving the resilience of new cultivars to these devastating diseases (Varshney et al., 2013). Additionally, the use of MAS in common bean has led to the successful pyramiding of multiple resistance genes, providing broad-spectrum resistance to various pathogens and pests. These examples underscore the potential of MAS to transform legume breeding by enabling the rapid and precise development of resistant cultivars, ultimately contributing to increased crop productivity and food security (Alkimim et al., 2017).

6 Genomic Selection (GS) in Legume Breeding for Resistance

6.1 Principles and applications of genomic selection

Genomic selection (GS) is a modern breeding approach that leverages genome-wide marker data to predict the breeding values of individuals, thereby facilitating the selection of superior genotypes. Unlike traditional marker-assisted selection, which focuses on identifying individual loci associated with traits, GS uses all available marker data to predict the overall genetic potential of an individual (Crossa et al., 2017; Budhlakoti et al., 2022).

This method is particularly advantageous for complex traits controlled by multiple genes with small effects, such as disease resistance in legumes (Budhlakoti et al., 2022). The application of GS in legume breeding has been bolstered by advances in next-generation sequencing and genotyping technologies, which have enabled the development of dense genetic maps and comprehensive genomic resources (Varshney et al., 2013).

6.2 Advantages of GS over traditional breeding methods

GS offers several advantages over traditional breeding methods. Firstly, it increases the accuracy of selection by using genome-wide markers, which capture more genetic variation than a few selected markers. This leads to more precise predictions of breeding values and, consequently, more effective selection of superior genotypes (Crossa et al., 2017). Secondly, GS accelerates the breeding cycle by allowing early selection of individuals based on their genomic estimated breeding values (GEBVs), thus reducing the need for extensive phenotyping (Budhlakoti et al., 2022). This is particularly beneficial for traits that are difficult to measure or are influenced by environmental factors, such as disease resistance (Carpenter et al., 2018). Additionally, GS can be integrated with other modern breeding approaches, such as marker-assisted backcrossing and genomic-enabled prediction, to further enhance the efficiency of breeding programs (Varshney et al., 2013; Bekele et al., 2019).

6.3 Genomic selection for pest and disease resistance in legumes: recent advancements

Recent advancements in GS have significantly improved the breeding of legumes for pest and disease resistance. For instance, GS has been successfully applied to improve resistance to ascochyta blight in pea, a disease that is challenging to assay due to its environmental dependency and the involvement of multiple pathogens. The use of GS models, such as genomic best linear unbiased prediction (GBLUP) and Bayesian methods, has shown promising results in predicting disease resistance with high accuracy (Carpenter et al., 2018). Similarly, GS has been employed to enhance resistance to Fusarium wilt and Ascochyta blight in chickpea, and late leaf spot and leaf rust in groundnut, demonstrating its potential to develop superior cultivars with enhanced tolerance to various diseases (Varshney et al., 2013).

Moreover, the integration of GS with other genomic tools, such as genome-wide association studies (GWAS) and high-throughput phenotyping, has further refined the selection process. For example, the combination of GS with hyperspectral imaging technology has been proposed to improve the accuracy of genomic predictions and accelerate the breeding cycle (Crossa et al., 2017). These advancements underscore the transformative potential of GS in legume breeding, paving the way for the development of disease-resistant varieties that can thrive in diverse and challenging environments (Kankanala et al., 2019; Jha et al., 2022).

7 Genetic Engineering and CRISPR-Cas9 in Resistance Breeding

7.1 Introduction to transgenic approaches and CRISPR-Cas9 technology

Genetic engineering has revolutionized the field of plant breeding, offering precise and efficient methods to enhance pest and disease resistance in crops. Traditional transgenic approaches involve the introduction of foreign genes into a plant's genome to confer desired traits. However, these methods often face regulatory hurdles and public resistance due to concerns over genetically modified organisms (GMOs). The advent of CRISPR-Cas9 technology has marked a significant advancement in genetic engineering. CRISPR-Cas9 allows for targeted genome editing by creating double-strand breaks at specific locations in the DNA, which are then repaired by the cell's natural mechanisms, leading to precise modifications. This technology is not only more efficient and cost-effective but also enables the development of transgene-free plants, addressing some of the concerns associated with traditional transgenic methods (Borrelli et al., 2018; Ahmad et al., 2020; Nascimento et al., 2023).

7.2 Engineering pest and disease resistance genes in legumes

The application of CRISPR-Cas9 in legumes has shown promising results in enhancing resistance to various pests and diseases. By targeting specific genes associated with susceptibility, researchers can create mutations that confer resistance. For instance, the CRISPR-Cas9 system has been used to disrupt the function of genes that facilitate viral infections, thereby developing virus-resistant plants (Chandrasekaran et al., 2016; Borrelli et al., 2018). In legumes, specific genes related to pest and disease resistance have been identified and targeted using CRISPR-Cas9. For example, the UDP-glycosyltransferase (UGT) gene in soybeans has been edited to enhance

resistance against leaf-chewing insects. Mutations in this gene resulted in increased flavonoid biosynthesis, which is associated with insect resistance (Zhang et al., 2022). Similarly, genes involved in the plant's defense mechanisms against fungal and bacterial pathogens have been modified to improve resistance (Borrelli et al., 2018; Erdoğan et al., 2023).

7.3 Case studies of successful genetic engineering efforts for resistance traits in legumes

A typical case study highlights the successful application of genetic engineering and CRISPR-Cas9 in developing pest and disease-resistant legumes. Using CRISPR-Cas9, researchers introduced targeted mutations in the *GmUGT* gene, resulting in soybean varieties with enhanced resistance to pests like *Helicoverpa armigera* and *Spodoptera litura*. This modification did not affect the overall plant phenotype, demonstrating the precision and effectiveness of CRISPR-Cas9 in pest resistance breeding (Figure 2) (Zhang et al., 2022). This case study underscores the transformative impact of CRISPR-Cas9 technology in resistance breeding, offering new avenues for developing robust legume varieties capable of withstanding biotic stresses.

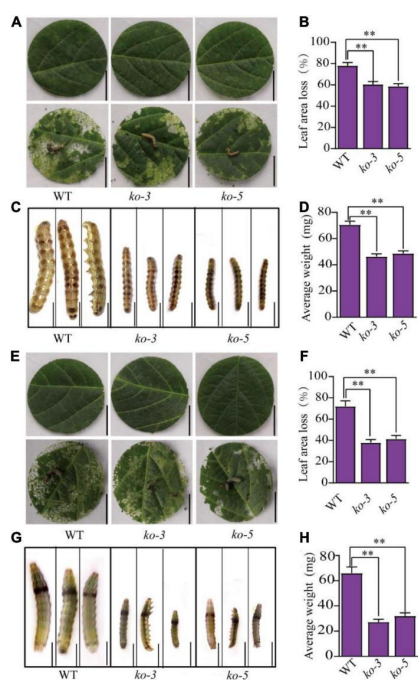


Figure 2 The CRISPR/Cas9-mediated mutagenesis of *GmUGT* enhanced resistance to *H. armigera* and *S. litura* in soybean (Adopted from Zhang et al., 2022)

Image caption: (A) The phenotype of detached leaves of *ko-3*, *ko-5*, and WT plants attacked by *H. armigera* for 3 days (bar = 2 cm). (B) Percentage of leaf area loss in *ko-3*, *ko-5*, and WT plants attacked by *H. armigera* for 3 days (n = 50 biological repeats). (C) The phenotype of *H. armigera* larvae fed detached leaves of *ko-3*, *ko-5*, and WT plants for 7 days (bar = 0.2 cm). (D) The average weight of *H. armigera* larvae that were fed detached leaves of *ko-3*, *ko-5*, and WT plants for 7 days (n = 50 larvae). (E) The phenotype of detached leaves of *ko-3*, *ko-5*, and WT that was attacked by *S. litura* for 3 days (bar = 2 cm). (F) Percentage of leaf area loss in *ko-3*, *ko-5*, and WT plants attacked by *S. litura* for 3 days (n = 50 biological repeats). (G) The phenotype of *S. litura* larvae that were fed detached leaves of *ko-3*, *ko-5*, and WT plants for 7 days (bar = 0.2 cm). (H) The average weight of *S. litura* larvae that were fed detached leaves of *ko-3*, *ko-5*, and WT for 7 days (n = 50 larvae). Data shown are means and standard deviations. Statistically significant differences are marked with asterisks (** $p < 0.01$; Student's *t*-test) (Adopted from Zhang et al., 2022)

8 Case Study: Molecular Breeding for Resistance to Fusarium Wilt in Chickpea

8.1 Overview of Fusarium wilt and its impact on chickpea production

Fusarium wilt, caused by the soil-borne fungus *Fusarium oxysporum* f. sp. *Ciceris* (FOC), is a major disease affecting chickpea (*Cicer arietinum* L.) production globally. This disease can lead to significant yield losses, sometimes up to 100% under severe conditions (Jha et al., 2020; Choudhary et al., 2022). Fusarium wilt is particularly problematic because it affects the vascular system of the plant, leading to wilting and eventual plant death. The disease is exacerbated by certain environmental conditions and is a persistent problem in many chickpea-growing regions (Mannur et al., 2018; Choudhary et al., 2022).

8.2 Application of molecular markers and genomic tools to develop wilt-resistant chickpea varieties

Recent advances in molecular breeding have significantly contributed to the development of *Fusarium* wilt-resistant chickpea varieties. The use of molecular markers, such as simple sequence repeats (SSRs) and single nucleotide polymorphisms (SNPs), has enabled the identification of quantitative trait loci (QTLs) associated with resistance to different races of *Fusarium* wilt (Sabbavarapu et al., 2013; Garg et al., 2018; Yadav et al., 2023). Marker-assisted backcrossing (MABC) has been employed to introgress resistance genes from donor parents into elite cultivars, resulting in the development of improved lines like Super Annigeri 1 and JG 74315-14 (Mannur et al., 2018). Additionally, genomic tools such as RNA-seq, proteomics, and metabolomics have provided deeper insights into the plant-pathogen interactions and the molecular basis of resistance (Figure 3) (Jha et al., 2020; Yadav et al., 2023).

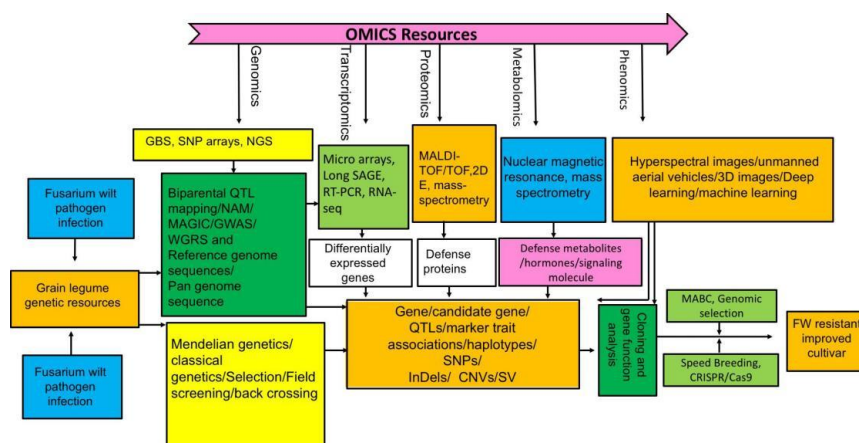


Figure 3 Integrated breeding, genetics, and “omics” scheme illustrating how to combat FW resistance in grain legume (Adopted from Jha et al., 2020)

8.3 Outcomes of the breeding program and lessons learned

The molecular breeding programs have yielded several successful outcomes. For instance, the development of Super Annigeri 1 and improved JG 74 lines demonstrated enhanced resistance to *Fusarium* wilt and increased yield performance (Mannur et al., 2018). The identification of specific QTLs and linked markers has facilitated the rapid development of resistant cultivars through marker-assisted selection (MAS) (Sabbavarapu et al., 2013; Garg et al., 2018). One of the key lessons learned is the importance of integrating multiple genomic approaches to understand the complex nature of disease resistance and to develop robust resistant varieties (Jha et al., 2020; Yadav et al., 2023). Additionally, the need for high-throughput phenotyping and the challenges associated with it have been highlighted as critical areas for future improvement (Jha et al., 2020).

8.4 Potential for scaling similar breeding strategies to other legume crops

The success of molecular breeding for *Fusarium* wilt resistance in chickpea provides a valuable framework that can be scaled to other legume crops. Similar strategies can be employed to tackle *Fusarium* wilt in other legumes such as faba bean and common bean, where molecular markers and genomic tools have already shown promise (Leitão et al., 2020; Mahmoud and El-Fatah, 2020). The principles of identifying resistance-associated QTLs, utilizing marker-assisted selection, and integrating omics approaches can be universally applied to enhance disease resistance across various legume species (Jha et al., 2020; Leitão et al., 2020; Mahmoud and El-Fatah, 2020). This approach not only improves crop resilience but also contributes to sustainable agricultural practices by reducing the reliance on chemical control measures.

9 Challenges and Limitations in Molecular Breeding for Resistance

9.1 Complex genetics of resistance traits in legumes

The genetic basis of resistance traits in legumes is often complex and polygenic, making it challenging to identify and manipulate these traits effectively. For instance, resistance to rust in legumes such as faba bean, pea, chickpea, and lentil is typically incomplete and governed by multiple quantitative trait loci (QTLs), which complicates

breeding efforts. Additionally, the large genome sizes of many legumes further hinder the development of comprehensive genomic resources and the saturation of genetic maps, which are essential for effective marker-assisted selection (MAS). The intricate interactions between legumes and their pathogens, as well as the involvement of multiple genes and pathways, add another layer of complexity to breeding for resistance (Kankanala et al., 2019).

9.2 Challenges in identifying and validating resistance genes

Identifying and validating resistance genes in legumes is a significant challenge due to the continuous evolution of pathogens and the dynamic nature of plant-pathogen interactions. The high selection pressure exerted by pathogens can lead to the breakdown of resistance genes in the field, necessitating ongoing efforts to discover and incorporate new resistance genes (Kankanala et al., 2019). Moreover, the integration of diverse genomic data, such as QTL mapping, genome-wide association studies (GWAS), and transcriptomics, is required to pinpoint key resistance genes and their regulatory networks (Jha et al., 2022; Jha et al., 2023). The complexity of these interactions and the need for high-throughput phenotyping and genotyping tools further complicate the identification and validation process (Rubiales et al., 2015; Saxena et al., 2023).

9.3 Socioeconomic and regulatory barriers to the adoption of molecular breeding technologies

The adoption of molecular breeding technologies in legumes is often hindered by socioeconomic and regulatory barriers. In many developing countries, where legumes are a staple crop, limited financial resources and infrastructure can impede the implementation of advanced breeding techniques (Jha et al., 2023). Additionally, regulatory frameworks governing the use of genetically modified organisms (GMOs) and other biotechnological interventions can be restrictive, slowing down the deployment of improved legume varieties. Public perception and acceptance of GMOs also play a crucial role, as consumer resistance to genetically engineered crops can influence policy decisions and market dynamics.

9.4 Environmental concerns and the potential for resistance breakdown

Environmental factors and the potential for resistance breakdown pose significant challenges in molecular breeding for legume resistance. The continuous interaction between legumes and their pathogens in diverse environmental conditions can lead to the emergence of new pathogen strains that overcome existing resistance genes (Wille et al., 2018; Kankanala et al., 2019). Climate change further exacerbates this issue by altering the distribution and prevalence of pests and diseases, necessitating the development of resilient legume varieties that can withstand these changes (Jha et al., 2022). Additionally, the potential for resistance breakdown requires breeders to adopt a holistic approach, considering the entire plant-microbiome interaction and incorporating beneficial microbes into breeding strategies to enhance plant health and resistance (Wille et al., 2018).

10 Future Directions and Opportunities in Molecular Breeding

10.1 Integration of molecular breeding with other emerging technologies

The integration of molecular breeding with emerging technologies such as genome editing and phenomics holds significant promise for enhancing pest and disease resistance in legumes. Genome editing tools, particularly CRISPR/Cas9, have demonstrated substantial potential in developing insect and pathogen-resistant crops by altering effector-target interactions, knocking out host-susceptibility genes, and engineering synthetic immune receptors (Bisht et al., 2019). Additionally, advancements in OMICs technologies, including transcriptomics, proteomics, and metabolomics, have revolutionized plant breeding by identifying key genes and pathways involved in stress responses, which can be targeted for genome editing (Ali et al., 2022; Jha et al., 2022). The combination of these technologies with high-throughput phenotyping tools can accelerate the breeding process and improve the precision of selecting desirable traits (Jha et al., 2022).

10.2 The role of molecular breeding in developing climate-resilient and resistant legume varieties

Molecular breeding plays a crucial role in developing climate-resilient and resistant legume varieties. The increasing incidence of diseases due to climate change necessitates the development of robust legume cultivars. Genomic resources, such as QTL mapping and genome-wide association studies, have identified resistance genes and genomic regions that can be introgressed into elite cultivars to enhance resistance to diseases like *Ascochyta*

blight and Fusarium wilt (Jha et al., 2020; Jha et al., 2022). Moreover, the integration of genomics-assisted breeding with traditional breeding methods has led to the development of climate-resilient legume varieties with improved yield and stress tolerance (Thudi et al., 2020; Roy et al., 2023). These advancements ensure the sustainability of legume production in the face of changing environmental conditions.

10.3 Policy recommendations to support molecular breeding initiatives

To support molecular breeding initiatives, several policy recommendations can be made. Governments and funding agencies should allocate more resources to research in molecular breeding and genomics to accelerate the development of resistant legume varieties (Shunmugam et al., 2018). Encourage collaboration between public and private sectors, and promote data sharing among researchers to enhance the efficiency of breeding programs (Duc et al., 2015). Develop clear and supportive regulatory frameworks for the use of genome editing technologies in crop improvement to facilitate the adoption of these technologies (Bisht et al., 2019). Invest in training programs to build the capacity of researchers and breeders in advanced molecular breeding techniques (Varshney et al., 2013). Increase public awareness about the benefits of molecular breeding and genome editing to gain public support and acceptance (Jha et al., 2023).

10.4 Future research priorities to enhance pest and disease resistance in legumes

Future research priorities to enhance pest and disease resistance in legumes should focus on the following areas. Conduct comprehensive functional genomics studies to identify and characterize genes involved in pest and disease resistance (Jha et al., 2020; Jha et al., 2023). Develop and refine genome editing techniques to create precise modifications in resistance genes and pathways (Bisht et al., 2019). Implement high-throughput phenotyping platforms to accurately assess resistance traits and accelerate the breeding process (Jha et al., 2022). Utilize integrated OMICS approaches to understand the complex interactions between legumes and pathogens, and identify novel resistance mechanisms (Ali et al., 2022; Jha et al., 2022). Create multi-parent advanced generation intercross (MAGIC) populations to increase genetic diversity and identify new resistance alleles (Varshney et al., 2013).

Focus on breeding strategies that combine resistance to pests and diseases with climate resilience to ensure sustainable legume production (Thudi et al., 2020; Roy et al., 2023). Explore synthetic biology approaches, such as synthetic promoters and transcription factors, to enhance resistance traits in legumes (Jha et al., 2023). Foster collaborative research efforts between breeders, geneticists, and pathologists to address the multifaceted challenges of pest and disease resistance (Shunmugam et al., 2018). By prioritizing these research areas, the scientific community can develop robust legume varieties that are well-equipped to withstand the challenges posed by pests, diseases, and climate change.

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Conflict of Interest Disclosure

The authors affirm that this research was conducted without any commercial or financial relationships that could be construed as a potential conflict of interest.

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