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Molecular Identification and Breeding Strategies of Rice Blast Resistance Genes

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Abstract The harm of rice blast and the importance of rice have always received widespread attention. This review aims to explore the molecular identification and breeding strategies of rice blast resistance genes, providing a detailed overview. It elaborates on the methods for identifying blast resistance genes, including past genetic research and the application of modern molecular biology technology, as well as molecular breeding strategies for blast resistance. It emphasizes the importance of molecular marker assisted breeding of resistant varieties. This review provides detailed information on molecular breeding methods, showcasing developed blast resistant rice varieties and their applications in different regions. This is one of the main results, combined with current challenges and future prospects, to help readers understand the future direction of this field. The article summarizes the importance of molecular identification of blast resistant genes in rice and molecular breeding strategies for global food security, And how to address future challenges, this topic provides a theoretical basis for future research and decision-making.

Keywords Wild rice (*Oryza rufipogon*); Genetic diversity; Adaptability; Rice breeding; Molecular identification

1 Introduction

Rice (*Oryza sativa*), as one of the world's most important food crops, plays a vital role in meeting global food demand. However, the stability and growth of rice yield are facing threats from a variety of biotic and abiotic factors. Among them, rice blast (*Magnaporthe oryzae*) is one of the diseases that poses a major threat to rice yield. This study aims to further explore the molecular identification and molecular breeding strategies of rice blast resistance genes, and contribute to the sustainability of global rice production and food security.

And one of the important diseases that harm rice. Early rice blast occurs in various places, especially ear blast that occurs at the ear neck node of rice and often causes "white ears", resulting in reduced early rice yields and even loss of harvest in some fields. In particular, high-quality early rice, cold-soaked fields and rice blast "old areas" are seriously affected. Because the early rice growing season, especially before and after heading, is a rainy period with high air humidity, which is conducive to the occurrence and damage of rice blast (Tembo et al., 2020).

For many years, scientific researchers have been working to identify and utilize genetic factors responsible for rice blast resistance to enhance rice's resistance to this disease. Early research relied mainly on genetic methods such as genetic linkage mapping and cross-breeding. However, with the advancement of molecular biology and genomics technology, scientists have been able to study the rice genome in depth, accelerating the identification of rice blast resistance genes. Molecular breeding strategies have also attracted increasing attention. Through molecular marker-assisted breeding, researchers can more accurately select individuals with disease resistance, thereby shortening the breeding cycle. This strategy not only improves the breeding efficiency of rice blast-resistant varieties, but is also expected to reduce dependence on chemical pesticides, thereby reducing environmental burden (Chukwu et al., 2019).

The study is to summarize key research results on the identification of rice blast resistance genes and molecular breeding strategies and to provide a comprehensive understanding of these strategies. Our intention is to highlight the importance of molecular characterization and molecular breeding in improving rice disease resistance and global food security. We will rely on existing research results to deeply analyze the potential of molecular

marker-assisted breeding and the advantages of combining it with traditional breeding methods. In this challenging era, we believe that through in-depth research and informed decision-making, we can further improve rice's resistance to rice blast and make an important contribution to the global food supply and farmers' livelihoods. At the same time, we will explore future research directions to promote the development of more disease-resistant rice varieties to ensure global food security.

2 Research Progress on Rice Blast Disease

2.1 Overview of rice blast disease

The pathogen of rice blast (*Magnaporthe oryzae*) is a fungus that is highly specialized and mainly infects rice plants, but can also cause similar diseases on other grass plants. The fungus overwinters on rice straw and rice with mycelium and conidia. When the temperature rises to about 20 °C the following year, a large number of conidia will be produced when rainfall occurs. Conidia are spread by air currents, raindrops, running water, and insects. In the presence of water and suitable temperature, appressoria germinate, produce hyphae, invade the host, and produce lesions. Under suitable temperature and humidity conditions, new conidia are produced, re-infected, and gradually expand and spread.

The optimum temperature for mycelial growth is 25 °C to 28 °C; the optimum temperature for spore formation is 26°C to 28°C, and a relative humidity of more than 90% is required. Focus on areas with less sunshine, long duration of fog and dew, mountainous areas and areas along rivers and coastal areas with mild climate. Suitable temperature and high humidity are conducive to the onset of disease. Rice is most susceptible to the disease from the 4-leaf stage to the tillering stage and heading stage. When rice is in the susceptible stage, the temperature is between 20 °C and 30 °C, especially between 24 °C and 28 °C, there are many rainy days, and the relative humidity remains above 90%, which can easily cause serious occurrence of rice blast. Excessive, partial or late application of nitrogen fertilizer is conducive to bacterial infection; long-term deep irrigation or cold water irrigation will aggravate the disease (Ning et al., 2020).

The life cycle of pathogenic bacteria includes multiple reproductive stages, the most important of which are conidia, ionic fruiting bodies and asexual spores. Conidia are the most common form of transmission of this pathogen, which are extremely small spores that are spread to rice leaves by wind. Asexual spores are another important form of transmission and are produced on infected tissues. It has a highly diverse genetics, and this diversity can develop through the evolution of different habitats and pathogenic bacterial strains, making it extremely adaptable. This is one of the reasons why this pathogen can cause disease in different geographical areas and rice varieties by producing special infection structures called "infectors" that penetrate the outer layer of rice leaves. Once inside the plant, it will infect different parts such as leaves, stems, and ears, leading to the spread of the disease. It has thousands of genes, some of which encode infection-related proteins, toxins and resistance genes. These genes play a key role in controlling the infection ability and resistance of pathogenic bacteria.

The genome contains many genes encoding pathogenicity-related proteins that encode toxins, components of infection structures, and proteins involved in host interactions. Among them, pathogenicity-related proteins are key factors in infecting rice. Different expression patterns and combinations of pathogenic genes can affect the infectivity of pathogenic bacteria, allowing them to infect different rice varieties and resistant genotypes (Spence et al., 2014).

The genetic diversity of pathogenic bacteria enables them to adapt to different rice varieties and environmental conditions, and there are extensive genetic differences between pathogenic bacteria strains, which is partly due to the high adaptability and evolutionary power of pathogenic bacteria. This diversity increases the complexity of the disease because different strains may produce different infectivity in resistant varieties, making it difficult to sustain resistance in resistant varieties.

The control of rice blast mainly relies on disease-resistant rice varieties. Therefore, it is crucial to understand the pathogenic mechanism of pathogenic bacteria and the genetic basis of disease-resistant rice. By further studying

the function and expression patterns of disease resistance genes, more disease-resistant rice varieties can be developed. This will not only help reduce the threat of diseases to global rice production, but also help reduce dependence on pesticides and reduce agricultural production costs.

2.2 Diversity and mutation mechanism of rice blast fungus

Rice blast is an important pathogenic fungus of rice, and its diversity and mutation mechanism pose a serious threat to rice production. When researching this area, we need to gain a deeper understanding of the diversity of pathogens and how they mutate in different environments. The diversity of *M. oryzae* is mainly reflected in the genetic differences between different strains. These differences can lead to different abilities of different pathogen strains to resist straw rice, thus affecting the yield and quality of rice.

For example, a strain of rice blast fungus discovered in Guangdong Province, China, has a relatively strong ability to fight rice. By analyzing the genome of this strain, scientists discovered a series of genes that are highly adapted to it, allowing it to infect rice plants more efficiently. This example highlights the real-world impact of *M. oryzae* diversity, as the emergence of this strain could lead to a threat to traditional rice varieties, requiring more resistant varieties to maintain agricultural production. The diversity of pathogenic fungi is also reflected in the variability of their life cycles. The rice blast fungus may adopt different life cycle strategies in different environments, which results in certain variability in its disease prevalence in different regions or seasons. For example, during dry seasons, *M. oryzae* may adopt a dormant life cycle to adapt to the dry conditions of the environment. And during the wet season, it may transform into a more invasive life cycle to infect rice plants more effectively (Nasruddin and Amin, 2013).

In terms of mutation mechanism, the genome of *M. oryzae* has certain variability, which provides a genetic basis for its adaptation to different environmental conditions. Genome variation can be achieved through a variety of ways, including genetic recombination, gene mutation, etc. For example, in some cases, genetic recombination may occur between two different strains of *M. oryzae*, resulting in subsequent strains with new genetic characteristics that make them better suited to specific environments. This mechanism of genome variation enables pathogenic fungi to adapt to different ecological conditions in a relatively short period of time, posing challenges to agricultural production. Another mechanism of variation is genetic mutation, where the gene itself changes randomly. This variation may cause the pathogen to exhibit new characteristics, sometimes new adaptations against resistant varieties. For example, some studies have found that certain strains of the rice blast fungus have acquired the ability to infect certain resistant varieties through genetic mutations. This mutation mechanism allows pathogenic fungi to quickly adapt to new resistant varieties in farmland, exacerbating the spread and damage of the disease (Chakraborty et al., 2021).

2.3 Molecular markers and mapping of rice blast resistance genes

Molecular markers and mapping of rice blast resistance genes are important means to improve rice disease resistance and cultivate disease-resistant varieties. By in-depth studying the distribution, genetic mechanism and diversity of resistance genes, scientists have provided more possibilities for future rice breeding work.

Rice blast caused by *Magnaporthe oryzae* is one of the most important fungal diseases of rice. The use of disease resistance genes to prevent and control this disease is the most economical, effective and environmentally friendly method. For example, in a study by the Academy of Agricultural Sciences, a new gene Pijx related to rice blast resistance was identified on chromosome 12 of rice. This gene has broad-spectrum resistance throughout the growth period; the Pijx protein interacts with the ATP synthase β subunit It interacts with the base and promotes its ubiquitination and degradation, thereby activating the activity of respiratory burst oxidase, inducing a burst of activated oxygen, and making the rice strain acquire resistance (Xiao et al., 2023) (Figure 1).

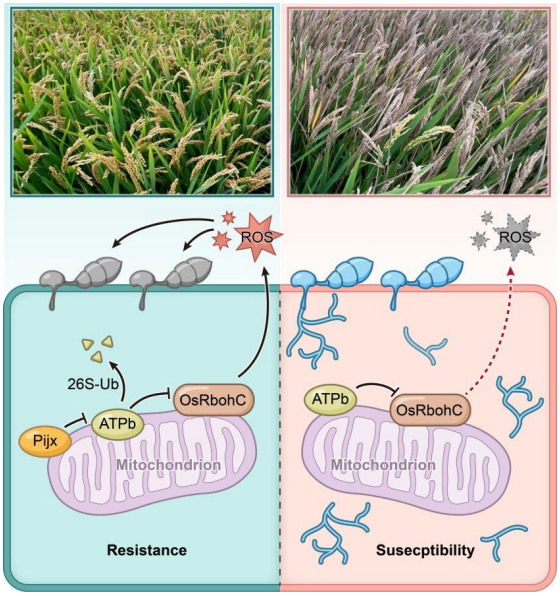

Figure 1 Molecular mechanism of Pijx gene regulating rice blast resistance (Xiao et al., 2023)

Molecular markers and mapping also help scientists gain a deeper understanding of the genetic mechanism of rice blast resistance genes. By locating these genes, scientists can study information such as their location in the rice genome and their interactions with other genes, which provides the basis for in-depth exploration of the rice blast resistance mechanism. For example, by locating rice blast resistance genes, scientists found that some resistance genes may be closely related to the plant's immune system, thus providing clues to further analyze the disease resistance mechanism of rice (Xiao et al., 2023).

Advances in molecular marker technology have also provided more possibilities for the discovery of rice blast resistance genes. With the development of high-throughput sequencing technology, scientists can analyze the rice genome more quickly and comprehensively, making it easier to discover genes related to rice blast resistance. For example, through whole-genome sequencing technology, scientists have successfully discovered some new rice blast resistance genes. The existence of these genes provides new candidate genes for breeding more disease-resistant rice varieties in the future

Molecular marker technology can also help analyze the diversity of rice blast resistance genes. Different rice varieties may carry different resistance genes, and the differences in these genes may be related to the geographical distribution of the varieties, ecological environment and other factors. By molecularly marking and locating the resistance genes of different varieties, scientists can map the distribution of rice blast resistance genes, reveal their diversity and distribution patterns, and provide a scientific basis for the rational selection of rice varieties suitable for different regions..

3 Molecular Identification of Rice Blast Resistance Genes

3.1 Application of genomics technology

The application of genomics technology in the agricultural field, especially whole-genome sequencing technology, provides a powerful tool for the discovery of resistance genes. Whole-genome sequencing is a high-throughput genetic analysis method that can quickly and accurately reveal the entire genome information of an organism. In the study of rice resistance genes, the application of whole-genome sequencing technology has made remarkable achievements.

An important application of whole-genome sequencing technology is in the discovery of resistance genes. By performing whole-genome sequencing of resistant varieties against rice blast, scientists are able to gain a comprehensive understanding of the composition of the rice genome and quickly identify genes associated with resistance. As mentioned before, by conducting whole-genome sequencing of a highly resistant rice variety,

researchers successfully discovered a new rice blast resistance gene *Pijx*. This gene was quickly identified through whole-genome sequencing, providing an important candidate gene for rice disease resistance breeding (Xiao et al. 2023).

Association analysis is another important method in genomics technology. It helps scientists find potential resistance genes by analyzing the correlation between genes and phenotypes. The advantage of association analysis is that multiple genes can be studied simultaneously in large-scale samples and the complex relationship between genes and phenotypes can be revealed. In rice disease resistance research, association analysis is widely used to find genetic variants associated with rice blast resistance.

Correlation analysis also has a wide range of advantages in practical applications. First, it can discover situations where multiple genes are simultaneously associated with disease resistance traits, which helps to fully understand the complexity of resistance mechanisms. Secondly, association analysis can also identify alleles that are beneficial to resistance, providing precise genetic information for subsequent molecular breeding. Through this method, scientists have achieved remarkable results in the discovery and utilization of rice blast resistance genes. In addition, association analysis technology can also help identify genomic regions associated with resistance, which provides a breakthrough for more in-depth molecular research.

3.2 Gene expression and functional studies

The study of gene expression and function is a key area for in-depth understanding of the genetic mechanism of organisms and the search for potential disease resistance genes. Differential expression analysis is an important method to study gene expression levels under different conditions. By comparing normal and pathogen-infected tissues, scientists were able to identify differentially expressed genes, providing important clues to the identification of disease-resistant genes.

The principle of differential expression analysis is to conduct extensive detection of the entire genome of an organism through high-throughput sequencing technology, such as RNA sequencing. This method not only measures the expression level of each gene, but also compares gene expression changes under different conditions. For example, after rice is infected by blast disease, a large amount of gene expression data can be obtained by sequencing RNA from infected and uninfected tissues. Then, through statistical analysis, genes that were significantly up-regulated or down-regulated in infected tissues were found, and these genes may be involved in the resistance response of rice to rice blast.

Pib, the first rice blast resistance gene, encodes a nucleotide-binding leucine-rich repeat (NLR) protein that mediates resistance to the avirulent gene *AvrPib* in *Magnaporthe oryzae*. By comparing the gene expression profiles of resistant varieties and infected varieties, the scientists found that the expression level of the *Pib* gene was significantly up-regulated in the resistant varieties. Further functional studies showed that *Pib* participates in the immune response of rice and enhances rice resistance to rice blast by regulating the expression of a series of immune-related genes. This example highlights the importance of differential expression analysis in the identification of disease resistance genes and provides a basis for in-depth understanding of disease resistance mechanisms (Peng et al., 2021). Functional genomics is a discipline that studies the function of genes in cells or entire organisms. Functional genomics plays a key role in the identification of disease resistance genes. By understanding the function of genes, scientists can delve deeper into the specific role of genes in disease resistance, thereby providing more targeted strategies for breeding and gene editing.

A typical application is to reveal the molecular mechanism of rice blast resistance genes through functional genomics. Taking the previously mentioned *Pib* gene as an example, through functional genomics, scientists found that *Pib* participates in the immune signaling pathway of rice and regulates multiple genes related to rice blast resistance. This meticulous functional analysis enables scientists to gain a more comprehensive understanding of the role of *Pib* genes in rice disease resistance and provides specific targets for future rice breeding efforts.

Functional genomics can also verify the function of candidate genes through methods such as gene silencing or overexpression. For example, the rice stress-resistant molecular breeding team used map-based cloning and CRISPR/Cas9 gene editing methods to prove that *Pib* in the rice variety "Yunyin" (YY) can effectively enhance its disease resistance. Further research found that SH3P2, a protein containing an SH3 domain, has a punctate structure with Pib, which is mainly co-localized at clathrin-coated vesicles in rice cells, and *SH3P2* can directly bind to the CC domain of *Pib* (Xie et al., 2022).

To further explore the function of *SH3P2* in *Pib* -mediated rice blast resistance, the researchers studied overexpression and CRISPR/Cas9 knockout transgenic lines and found that *SH3P2* itself has no basal resistance, but SH3P2 in YY Overexpression of impairs Pib-mediated resistance to AvrPib-bearing blast strains and cell death induced by Pib-AvrPib specific recognition. Further competition experiments revealed that SH3P2 can inhibit the self-association of the Pib-CC domain. It is shown that *SH3P2* is dependent on the *Pib* -mediated immune pathway and affects the formation of the smallest functional unit of Pib dimer by inhibiting the self-association of the CC domain, thereby negatively regulating Pib-AvrPib recognition-mediated resistance (Xie et al., 2022).

3.3 Mapping of rice blast resistance QTL

Rice blast resistance QTL (Quantitative trait loci) is an important research work. Through this work, scientists can discover the genetic factors that control rice blast resistance traits and provide important information for breeding. The steps of quantitative trait locus analysis mainly include multiple stages such as population establishment, phenotypic and genotypic data collection, and QTL analysis. The application of this method provides a powerful tool for rice disease resistance breeding. Combined with the practical significance of molecular marker-assisted breeding, it lays the foundation for the cultivation of highly efficient disease-resistant rice varieties.

Mapping rice blast resistance QTL requires establishing a population with rich genetic variation. Typically, scientists choose to cross parents with different disease resistance traits to obtain a series of offspring, forming a genetically diverse population. Taking the cross between a certain resistant rice variety and a susceptible variety as an example, through large-scale phenotypic observation of the hybrid progeny, data on resistance traits are collected and a population suitable for QTL analysis is established (Peng et al., 2021).

Scientists need to collect a large amount of molecular marker data, including single nucleotide polymorphism (SNP) markers, parsimonious sequence repeat (SSR) markers, etc. These molecular markers can represent different regions in the rice genome. By analyzing molecular markers for each individual in the population, a high-density genetic map can be constructed. This step helps determine the genotypes of different individuals in the rice population and provides data support for QTL analysis.

When conducting QTL analysis, scientists usually use statistical methods, such as association analysis or population genetic analysis, to identify QTL related to rice blast resistance. By comparing individuals in a population showing different disease resistance traits and combining their genotype information, a series of QTL that may be related to resistance can be found. The location information of these QTL helps understand the genetic basis of rice blast resistance traits and provides a strong genetic basis for rice disease resistance breeding (Singh et al., 2021).

Molecular marker-assisted breeding has practical significance in disease resistance breeding. Through the use of molecular markers, breeders can screen plants with target disease resistance QTL at an early stage. This helps improve breeding efficiency and reduce waste of resources and time. Taking the previous example as an example, if the disease resistance QTL of a rice variety has been determined through QTL analysis, breeders can use molecular marker technology to directly screen individuals carrying the target QTL in the hybrid progeny to quickly form germplasm resources with resistance genes.

Molecular marker-assisted breeding can also help breeders conduct precise genomic selection. Through molecular marker technology, breeders can conduct targeted selection of specific regions in the rice genome, which not only

improves the transmission of disease resistance genes efficiency, and also avoids the carriage of undesirable genes unrelated to the target traits. This provides a more precise genetic improvement method for breeding more resistant rice varieties.

4 Molecular Breeding Strategies

4.1 Selection based on molecular markers

Molecular marker -assisted selection (MAS) is an indirect selection using molecular markers that are closely linked to the target trait gene. It is a selection of the target trait at the DNA level and is not affected by the environment or alleles. The interference of dominant and recessive relationships makes the selection results reliable, and at the same time it can avoid the interference of dominant and recessive relationships between alleles, thereby achieving efficient improvement of comprehensive traits such as crop yield, quality and resistance; marker-assisted breeding with marker genotype identification can It can be carried out in low generations and at any stage of plant growth. Co-dominant molecular markers allow the identification of recessive genes at the hybrid stage. The selection of target genes has the advantage of not being affected by gene expression and environmental conditions.

Bergman et al. (2000) of the State Key Laboratory of Crop Genetic Improvement of Huazhong Agricultural University used MAS technology to successfully introduce the broad-spectrum bacterial blight resistance gene Xar21 into the excellent restorer line Minghui 63, and selected the genes to be less than 1.0 cM on both sides of Xcr21 Through a round of backcrossing and selfing, single plants homozygous for Minghui 63 alleles at most loci except Xar21 were obtained. It can be seen that as long as there are appropriate molecular markers, MAS can indeed improve the efficiency of resistance selection and speed up the process of disease resistance breeding.

In addition to MAS technology, haploid selection (DH) breeding has made important progress in both theoretical and applied research. Through rice haploid breeding, we can speed up the breeding process, improve breeding efficiency, and provide new ways for rice breeding. At the same time, it can also reveal the genetic patterns and gene functions of rice, providing a basis for genetic improvement and functional gene research in rice. Some results have also been achieved in the practical application of rice blast. For example, haploid hybridization and cytology techniques are used to develop new rice varieties with disease resistance, stress tolerance and high yield.

Haploid selection (DH) is a breeding method that accelerates gene fixation by halving the number of chromosomes in a plant's cells to obtain homozygous haploid plants. In disease resistance breeding, DH technology can accelerate the fixation of genotypes carrying disease resistance genes, thereby faster obtaining new varieties with stable disease resistance traits (Peng et al., 2021).

Taking the fixation of rice blast resistance genes as an example, breeders first identify and select hybrid plants carrying rice blast resistance QTL through methods such as marker-assisted selection. Then, haploid selection technology is used to produce haploid offspring from these plants. Because a haploid plant is homozygous, all its cells carry the same genotype, including the target disease resistance gene. Through haploid selection, breeders can more quickly fix rice blast resistance genes in the entire plant population to form haploid germplasm with disease resistance traits.

The combined application of these two technologies also shows great potential. By combining marker-assisted selection with haploid selection, breeders can accurately select plants carrying target disease-resistant genes while accelerating the fixation of disease-resistant genes. This not only improves breeding efficiency, but also ensures the stable transmission of disease-resistant genes, providing a more reliable way to cultivate crop varieties with stronger disease resistance.

4.2 Application of gene editing technology

The basic principle of CRISPR-Cas9 technology involves CRISPR (Clustered regularly interspaced short palindromic repeats) sequences and Cas9 proteins. CRISPR sequences are a naturally occurring immune

mechanism in bacteria and archaea that record information about previously infected viruses or foreign DNA fragments. The Cas9 protein is a lipoendonuclease that can recognize and cut foreign DNA that matches the CRISPR sequence. During the gene editing process, scientists first design an RNA sequence that matches the DNA sequence of the target gene. This RNA sequence is guided by the CRISPR sequence and combines with the Cas9 protein to form a CRISPR-Cas9 complex. Once the complex matches the DNA sequence of the target gene, the Cas9 protein cuts the DNA sequence, causing the gene to be knocked out or added.

In terms of gene knockout, CRISPR-Cas9 technology is widely used in the study of rice disease resistance-related genes. For example, by designing an RNA sequence that matched a specific rice blast resistance gene, scientists successfully used CRISPR-Cas9 technology to knock out the gene. This method provides an effective means to study the role of specific disease resistance genes in rice disease resistance mechanisms. By observing the performance of rice plants after knockout, scientists can gain a deeper understanding of the impact of this gene on rice disease resistance traits (Romero and Gatica-Arias, 2019).

On the contrary, gene addition is another application direction of CRISPR-Cas9 technology, especially in the introduction of foreign genes to improve rice disease resistance. For example, scientists could engineer an RNA sequence to match a disease-resistant gene from another crop or plant. By using CRISPR-Cas9 technology, this RNA sequence is introduced into the rice genome to add exogenous disease resistance genes. This method provides an effective means for rice breeding to enhance disease resistance of rice by introducing exogenous genes. Taking the improved resistance of rice to rice stripe rust as an example, scientists have successfully transferred specific resistance genes from other plants to the rice genome through CRISPR-Cas9 technology. The addition of this foreign gene makes the rice plants more resistant to rice stripe rust. This method of gene addition provides an innovative approach to rice disease resistance breeding by introducing exogenous disease resistance genes to improve rice immunity to specific diseases.

Although CRISPR-Cas9 technology shows great potential in rice disease resistance research, there are still some potential problems that need to be paid attention to in practical applications, such as non-specific modification and incomplete gene editing. Nonspecific modifications may lead to undesirable side effects, while incomplete gene editing may produce polymorphic genotypes. Therefore, during the application of CRISPR-Cas9 technology, careful design and verification are required to ensure that the resulting rice varieties have the expected disease resistance traits.

4.3 Introduction of disease resistance genes

Transgenic technology is a method of introducing foreign genes into the cells of a target organism by changing the genetic material of the organism, thereby giving it new traits or functions. In the plant field, especially in crop breeding, transgenic technology is widely used to improve disease resistance. This study will outline the basic principles of transgenic technology and use examples to evaluate the effect of introducing disease resistance genes from other plants or microorganisms on plant disease resistance.

The basic principle of transgenic technology includes constructing a vector of foreign genes, introducing it into the cells of the target plant, and then integrating the foreign gene into the genome of the target plant through appropriate selection and screening methods. Among them, the most commonly used vector is Agrobacterium tumefaciens, which can introduce foreign genes into plant cells and integrate them into the plant genome. In addition, direct transgenic methods also include biological particle gun methods.

In terms of introducing disease resistance genes from other plants or microorganisms, a typical example is the introduction of exogenous disease resistance genes into rice. Taking the improved resistance of rice to rice blast as an example, scientists have successfully introduced the rice blast resistance gene *Xa21* through transgenic technology. This gene is derived from a naturally disease-resistant variety of rice and has strong resistance to rice blast. By introducing the *Xa21* gene into other rice varieties, we hope to improve the resistance of these rice varieties to rice blast (Sham et al., 2020).

The process of effect evaluation involves comprehensive observation and analysis of the disease resistance of transgenic rice. By comparing transgenic rice with wild-type rice or a control group, scientists can evaluate the impact of introducing the *Xa21* gene on rice disease resistance. Experiments have shown that transgenic rice shows stronger disease resistance when infected with rice blast, and its disease resistance is significantly improved compared to wild-type rice. This shows that the introduction of disease resistance genes from other plants can effectively enhance the disease resistance of rice, providing a feasible method to deal with disease threats (María et al., 2022).

There are some successful cases of introducing disease resistance genes from other plants or microorganisms to make plants show stronger immunity to specific pathogens. For example, attempts have been made to introduce antifungal genes from Arabidopsis into wheat to improve wheat resistance to fungal diseases. Through transgenic technology, the successfully introduced antifungal genes enable wheat to show a more powerful immune response when infected by fungi, thus reducing the impact of diseases on wheat. However, the application of genetically modified technology still faces some controversies and challenges, including ecological safety, food safety and other issues. Therefore, when introducing disease resistance genes from other plants or microorganisms, the overall performance of the plant needs to be carefully evaluated to ensure that it does not cause other adverse effects while resisting disease.

5 Promotion and Application

Field trials are an indispensable part of plant disease resistance breeding. By evaluating disease-resistant rice in actual planting environments, scientists can gain a more comprehensive understanding of its disease resistance, growth characteristics, and adaptability. This study will examine the evaluation of disease-resistant rice in field trials and explore the challenges and opportunities faced by field trials.

The evaluation of the effect of disease-resistant rice in actual planting environments is one of the core contents of field trials. By planting disease-resistant rice under real field conditions, scientists can observe its growth status, disease-resistant traits, and interactions with the natural environment. Interaction with surrounding plants. For example, in rice varieties introduced with rice blast resistance genes, scientists will plant them at field trial sites in different geographical locations to observe whether the disease-resistant rice exhibits strong resistance to rice blast in actual rice field environments (Cheng et al., 2021).

In a field trial of disease-resistant rice, scientists successfully introduced a rice blast resistance gene and planted it in multiple field trial sites. The results showed that these disease-resistant rice varieties showed excellent resistance to rice blast in different regions, and their yield losses were significantly reduced compared with traditional varieties. This field effect evaluation provides strong support for the application of disease-resistant rice varieties in actual farmland planting.

However, field trials also face a series of challenges. First, the complexity of the actual planting environment makes the experimental results more variable. Differences in soil, climate, pathogen pressure and other factors in different regions may affect the performance of disease-resistant rice. For example, the resistance of disease-resistant rice to rice blast may vary under different climate conditions, and trials need to be conducted at multiple locations to obtain a more comprehensive assessment (Eseola et al., 2021). Long-term observation and data collection are also a challenge. Plants have a relatively long growth cycle, especially crops such as rice, which take several months to harvest. During this process, environmental conditions, disease pressure and other factors may change, affecting the test results. In order to more comprehensively evaluate the effect of disease-resistant rice, scientists need to spend a long period of observation and data collection to ensure the reliability of the test results.

Field trials also present opportunities, particularly in validating the feasibility of laboratory findings in real-world settings. Field trials provide a realistic, complex farm environment that better simulates plant growth and interactions under natural conditions. For example, disease-resistant rice may face more ecological interactions in

actual farmland, including interactions with other organisms, pressure from natural pathogens, etc., and these factors can be fully considered in field trials (Li et al., 2019).

6 Outlook

Molecular identification and molecular breeding of rice blast resistance genes are of great significance in modern agriculture. With the increase in global food demand, the production of rice, one of the major food crops, is threatened by various diseases, among which rice blast is one of the main diseases affecting rice yield and quality. Therefore, in-depth research on rice blast resistance genes and their application in molecular breeding are of great strategic significance for improving rice disease resistance and ensuring food security. The achievements and discoveries that have been made have laid the foundation for the molecular identification and molecular breeding of rice blast resistance genes. Through the development of genomics technology, scientists have successfully identified a series of genes related to rice blast resistance, such as *Xa21*. The discovery of these genes enables us to gain an in-depth understanding of the genetic mechanism of rice blast resistance and provides strong support for further research and breeding efforts.

Future research directions include further exploring and identifying new rice blast resistance genes, and in-depth analysis of the specific mechanisms of these genes in the rice disease resistance process. With the development of high-throughput sequencing technology, we can more accurately analyze the sites related to rice blast resistance in the rice genome, thereby discovering more disease-resistant genes. In addition, researchers can also use functional genomics and other methods to conduct in-depth studies on the interaction between these genes and rice blast pathogens to provide more information for precision breeding.

The possible application of new technologies and methods is key to advancing genetic research and molecular breeding for rice blast resistance. Gene editing technology based on CRISPR-Cas9 provides a powerful tool for precise modification of the rice genome. By using CRISPR-Cas9 technology, scientists can directly perform site-specific editing of rice genes to enhance or suppress specific genes. This provides a way to optimize disease resistance genes and improve rice disease resistance. In addition, the application of single-cell sequencing technology allows us to gain a deeper understanding of the changes at each cell level in the rice disease resistance mechanism, providing detailed information for revealing the functional mechanism of rice blast resistance genes.

As research deepens, molecular breeding will play an even more important role in the future. By using technical means such as molecular marker-assisted breeding, genome selection, and gene editing, breeders can more accurately select and optimize rice genes and achieve efficient utilization of rice blast resistance genes. Molecular breeding can not only improve breeding efficiency, but also avoid some uncontrollable factors in traditional breeding, providing a more feasible way to breed more disease-resistant rice varieties. In the context of coping with climate change and global disease epidemics, molecular identification and molecular breeding of rice blast resistance genes will become important means to ensure food security. Through continuous in-depth research, we can expect to see the emergence of more efficient and stress-tolerant rice varieties resistant to rice blast in the future, making greater contributions to global food production.

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Conflict of Interest Disclosure

The author affirms 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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