



## Development of Alternaria Blight-Tolerant Lines of Indian Mustard (*Brassica juncea* L.): Emerging Strategies for Disease Resistance

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*Brassica juncea* L., (Indian mustard), is an important edible oilseed crop in India. Yield reduction due to biotic stresses is a major constraint in Indian mustard production. Alternaria blight caused by *Alternaria brassicae* is one of the major diseases of Indian mustard that has limited resistant cultivar in India. In Rabi (winter season) to isolate Alternaria blight-tolerant mutants in Indian mustard using gamma radiation and EMS mutagens. Morphologically and economically superior mutants of *Brassica juncea* were experimentally screened against Alternaria blight at the cotyledonary and adult stages. Mutants showing fewer conidia in infected cotyledons, exhibited tolerance to Alternaria blight. These mutants exhibited greater antioxidant capacity, and served as a defence against pathogens. Mutants showed lower levels of total soluble sugar content compared to susceptible cultivar Rohini, phenol content was higher, which may be responsible for resistance. Mutants appeared to be more resistant to Alternaria.

### Introduction

Oilseed crops constitute an important component of India's agricultural economy. A significant portion of the nation's oil production comes from brassicas. Among the *Brassica* species, Indian mustard (*Brassica juncea*) occupies the largest cultivated area in India. During the winter months from October to March, Indian mustard is widely cultivated in relatively drier cooler and relatively drier climates. The crop is mostly grown under rainfed conditions and is suitable for semi-arid to desert regions. In the Indian subcontinent, it is mainly cultivated for edible oil, leafy vegetables, sauces, condiments and high-protein livestock feed. States like Rajasthan, Haryana, Madhya Pradesh, Uttar Pradesh, Gujarat, and Punjab are major mustard producing states in India, while the crop is also grown in the northeastern region, including Assam and West Bengal (ICAR-IIRMR). Although India is already self-sufficient in many agricultural commodities, the country still imports a significant quantity of edible oil (Jat *et al.*, 2019). Therefore, increasing the productivity of mustard has become an important focus of agricultural research.

### Novel approaches for managing Alternaria blight disease through susceptibility (S) genes

Susceptibility genes contribute to the plant's vulnerability during pathogen infection. Modification or knockout these genes can reduce the success of pathogen invasion, thereby increasing tolerance or resistance to diseases. Targeting susceptibility genes using genome editing tools such as *CRISPR/Cas9* can facilitate the precise knockouts, knockdowns, or modifications of host susceptibility genes, which may durable and broad-spectrum resistance in *Brassica* crops (Ton *et al.*, 2025). The effectiveness of this strategy had been demonstrated through *CRISPR/Cas9* mediated editing of *eIF(iso)4E* gene in *Brassica rapa*, which conferred

resistance to Turnip Mosaic Virus (TuMV), an important viral pathogen (Lee *et al.* 2023). Similarly, *CRISPR/Cas9*-mediated modification of the host target gene *BnQCR8*, which interacts with the *Sclerotinia* effector *SsSSVPI*, increased resistance to both *Sclerotinia* and *Botrytis* (Zhang *et al.*, 2021). Identification of *Alternaria brassicae* susceptibility genes through transcriptomics, proteomics, or genetic approaches may provide new opportunities to improve disease resistance in Brassica crops using genome-editing technology.

### **Disease screening inoculum preparation for *Alternaria* blight**

In studies on *Alternaria* blight of *Brassica* crops, disease screening is commonly conducted under field conditions using artificial inoculation techniques. Seeds are generally sown during the appropriate growing season and standard disease screening procedures are followed as described in earlier studies.

### **Investigating Non-Host Resistance to Diseases Caused by *Alternaria***

Non-host resistance (NHR) refers to the natural ability of an entire plant species to naturally defend itself by all strains of a particular pathogen. This type of resistance is generally durable and broad-spectrum, making it an important strategy for developing disease-resistant crops.

### **Investigating RNAi technology**

Farmers are encouraged to adopt sustainable disease management practices such as crop rotation, biological control and the use of resistant varieties, in order to reduce dependence on chemical pesticides. However, excessive use of agrochemicals has resulted in the emergence of pesticide-resistant pathogens and the development of new disease strains, a problem that is further intensified by climate change. These challenges emphasize the need for innovative and long-lasting crop protection strategies.

Recent advances in biotechnology have introduced RNA interference (RNAi) as a promising strategy for managing plant diseases. RNAi is a natural biological mechanism that can silence or suppress specific genes in pathogens. Scientists are exploring this technology as a method to control plant diseases by targeting genes that are essential for the growth and infection ability of pathogens.

Two major RNAi-based approaches are currently being explored for crop protection. The first is Spray-Induced Gene Silencing (SIGS). In this method, artificially synthesized dsRNA molecules are sprayed onto plant surfaces. These molecules target essential genes in the pathogen and suppress its growth and infection process. SIGS is particularly attractive because it does not require the development of genetically modified crops, which may help reduce regulatory concerns and make the technology easier to adopt in agricultural systems. The second approach is HIGS (Host-Induced Gene Silencing), where plants are genetically engineered to produce small RNA molecules that can silence important genes in invading pathogens. When the pathogen infects the plant, these molecules interfere with the pathogen's gene expression and reduce its ability to cause disease.

The success of RNAi-based disease control depends on several factors, including the stability of dsRNA molecules, efficient delivery to the pathogen and the selection of suitable target genes. With the availability of genomic data for *Alternaria brassicae*, *Alternaria brassicicola*, scientists are now able to identify genes involved in toxin production, cell wall degradation, and other processes essential for pathogen survival (Dang *et al.*, 2015; Rajarammohan *et al.*, 2019). Targeting these genes using RNAi technology can help limit disease development.

### **Conclusion**

*Alternaria* blight remains one of the most serious diseases affecting Indian mustard production in India, leading to significant yield losses and limited availability of resistant cultivars. The development of *Alternaria* blight-tolerant lines through mutation breeding using gamma radiation and EMS has shown promising results. The tolerant lines were

characterized by lower conidia production, higher phenol content and increased antioxidant activity, which may contribute to enhanced defence mechanisms against the pathogen.

In addition to mutation breeding, modern biotechnological approaches such as genome editing, RNA interference (RNAi), and molecular breeding are providing new opportunities for improving disease resistance in Brassica crops. Techniques such as Spray-Induced Gene Silencing (SIGS) and Host-Induced Gene Silencing (HIGS) offer innovative strategies to target essential pathogen genes and reduce disease severity.

The integration of mutation breeding, molecular tools, and advanced biotechnological approaches can significantly accelerate the development of *Alternaria* blight-resistant cultivars of Indian mustard. Such strategies will play an important role in enhancing mustard productivity, reducing dependence on chemical pesticides, and ensuring sustainable oilseed production in India.

## References

1. Dang, H.X., Pryor, B., Peever, T. et al. 2015. The *Alternaria* genomes database: a comprehensive resource for a fungal genus comprised of saprophytes, plant pathogens, and allergenic species. *BMC Genomics* 16: 239. <https://doi.org/10.1186/s12864-015-1430-7>.
2. Jat, R.S., Singh, V.V., Sharma, P., Rai, P.K. 2019. Oilseed brassica in India: Demand, supply, policy perspective and future potential. *Oilseeds Fats, Crops and Oils* 26: 8.
3. Rajarammohan, S, Paritosh, K, Pental, D., Kaur, J. 2019. Comparative genomics of *Alternaria* species provides insights into the pathogenic lifestyle of *Alternaria brassicae* - a pathogen of the Brassicaceae family. *BMC Genomics* 20: 1036.
4. Ton, L.B., Qayyum, Z., Amas, J., Thomas, W.J.W., Edwards, D., Batley, J., Dolatabadian, A. 2025. Applications of *CRISPR/Cas* tools in improving stress tolerance in Brassica crops. *Front. Plant Sci.* 16:1616526. <https://doi.org/10.3389/fpls.2025.1616526>
5. Zhang, X., Cheng, J., Lin, Y., Fu, Y., Xie, J., Li, B., Bian, X., Feng, Y., Liang, W., Tang, Q., Zhang, H., Liu, X., Zhang, Y., Liu, C., Jiang, D. 2021. Editing homologous copies of an essential gene affords crop resistance against two cosmopolitan necrotrophic pathogens. *Plant Biotechnol. J.* 19: 2349-2361. [10.1111/pbi.13667](https://doi.org/10.1111/pbi.13667).