

**A seminar paper**  
**on**  
**Understanding Panicle Blast Resistance in Wheat: The Road Behind and The Road Ahead**

**Submitted to**

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# Understanding Panicle Blast Resistance in Wheat: The Road Behind and The Road Ahead<sup>1</sup>

By

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## ABSTRACT

Wheat blast is a devastating fungal disease of wheat caused by *Magnaporthe oryzae Triticum* that threatens global food security. The disease was first reported in Brazil in 1985 and has since spread to Bolivia, Paraguay, northern Argentina, the USA, Bangladesh, India, and Zambia. Farmers rely on cultural practices and fungicides, but overuse of fungicides led to fungal resistance, highlighting the need for blast-resistant genes to combat wheat blast. Several QTLs associated with wheat blast resistance have been identified, with the 2NS translocation being the most effective resistance locus. However, as 2NS translocation has recently been eroded, *Rmg8* and *RmgGR119* are the only effective and temperature-insensitive genes for both seedling and head stage resistance against wheat blast. Additionally, it is crucial to thoroughly test the effectiveness of the identified solutions under varying environmental conditions and particularly at the heading stage, as resistance at the seedling stage may not correspond with that at the adult stage. Various promising approaches can be utilized to create durable and robust blast-resistant varieties, including introgression of resistant genes into local cultivars, mutation breeding, genome editing approaches, and biological control. The review paper aims to explore existing knowledge for the effective deployment of achieve durable panicle blast resistance in wheat and offers some promising solutions to combat the wheat blast problem, whose successful implementation could result in improved crop yields and enhanced food security for farmers.

**Keywords:** panicle blast, *Magnaporthe oryzae Triticum*, gene introgression, genome editing

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## CHAPTER I

### INTRODUCTION

Wheat blast is a devastating fungal disease caused by the fungus *Magnaporthe oryzae* pathotype *Triticum* (MoT). This pathogen can infect wheat plants at any stage of growth, from seedling to maturity, and attacks the wheat plant's stem, leaves, and grain, causing significant yield losses and quality deterioration (Kohli *et al.*, 2011). It came into the scene in Bangladesh for the first time in 2016 and devastated 15,000 hectares of wheat cultivated area across eight districts in the southwestern part of Bangladesh with yield losses of up to 100% (Islam *et al.*, 2016). It was initially reported in Brazil and since then it has spread to South Asia and Africa (Das, 2017; Islam *et al.*, 2016; Igarashi, 1986; Tembo *et al.*, 2020).

Wheat blast is a fast-acting and devastating fungal disease and it threatens the food safety of tropical areas in South America, South Asia, and Africa. It directly strikes the wheat ear and can shrivel and deform the grain in less than seven days from the initial symptoms which gives farmers no time to act. The head infection results in complete or partial bleaching of the spike above the point of infection with either no grain or shriveled grain with low test weight (Islam *et al.*, 2016). The spike blast is seen developing in fields without visual symptoms of leaf blast. Controlling spike blast through the application of fungicides can be difficult as farmers cannot always see lesions on leaves before the heading stage to warrant fungicide application (Islam *et al.*, 2020).

Wheat is a crucial food source for 40% of the world's population and its consumption has particularly increased in South Asia and Sub-Saharan Africa (B.C. Curtis, 2022; Mottaleb *et al.*, 2018). With the projected increase in global population and food demand, wheat production must be safeguarded. This devastating disease significantly threatens to global food security, as it not only causes significant damage to wheat crops but also has the potential to spread to new wheat-growing regions through air and seed (Islam *et al.*, 2020; Kamoun *et al.*, 2019).

Effective management of this disease is a challenging task, as a singular approach is not sufficient to control it (Cruz & Valent, 2017). This highly destructive disease that progresses rapidly, leaving farmers with little time to prevent it (Islam *et al.*, 2019b). While chemical control methods can be ineffective under high disease pressure, fungicide resistance has been reported in Brazil (Castroagudín *et al.*, 2015). The most preferred and sustainable method for disease management is the use of blast-resistant wheat cultivars (Cruz & Valent, 2017). To date there are 10 blast-resistant genes and one blast-resistant chromosomal segment identified, however, most of them are temperature-labile and do not confer resistance at the adult or

heading stage (Cruppe *et al.*, 2020). There are only two promising bona fide blast-resistant genes, *Rmg8* and *RmgGR119*, however, they have not yet been cloned and characterized.

Breeding to achieve resistance against wheat blast has been a difficult task due to the complex genetics of resistance, coupled with the fungus's ability to rapidly evolve new races, thereby overcoming previously resistant varieties (Khan *et al.*, 2022). Thus, identifying new resistance genes is crucial to improve wheat blast resistance, considering the limited number of known resistance genes for the disease, and mutations can aid in this screening process. Furthermore, several secondary metabolites have also been identified by researchers as effective against MoT, with the potential to develop a novel biofungicide (Chakraborty *et al.*, 2022; Chakraborty *et al.*, 2020; Paul *et al.*, 2022; Rabby *et al.*, 2022).

Recent advancements in genomic approaches and bioinformatics tools can also aid in developing blast-resistant wheat varieties with greater accuracy and precision within limited time, making these approaches both ecologically and economically sustainable (Islam *et al.*, 2022). Additionally, it is important to note that blast resistance at the seedling stage is not consistent with that of the heading stage (Cruz *et al.*, 2012), and as wheat blast is predominantly a spike disease, greater attention should be given to assessing head blast or panicle blast.

Considering the aforementioned background, this seminar paper has been designed to fulfill the following objectives-

- i. To update the advancement of the understanding of the panicle blast
- ii. To review the current knowledge of wheat blast resistance
- iii. To explore the deployment of current knowledge for durable plant resistance

## **CHAPTER II**

### **MATERIALS AND METHODS**

The primary objective of this seminar paper is to conduct a review, and as a result, the majority of the data and information presented in this paper are collected from secondary sources. These sources consist of various published papers and Ph.D. dissertations, journals, books, articles, as well as online databases.

The constructive criticism and valuable suggestions provided by my major professor and course instructors were instrumental in improving the quality of this paper. Subsequently, the collected information was organized systematically and presented in this paper.



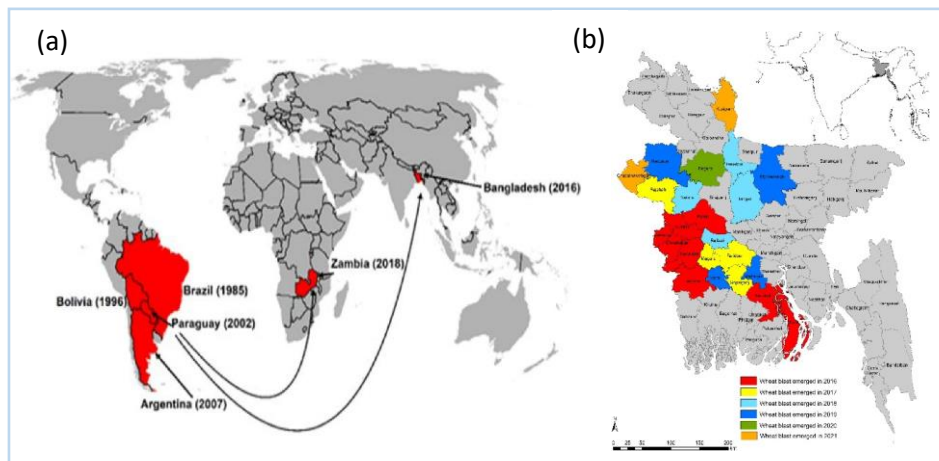
## CHAPTER III

### REVIEW OF FINDINGS

#### 3.1 Current knowledge of wheat blast

##### 3.1.1 Epidemiology

The first emergence of the deadly wheat blast disease was reported in the Brazilian state of Parana in 1985 (S. Igarashi, 1986), and it subsequently spread to the humid and warmer regions of Bolivia, Paraguay, and northern Argentina (Castroagudín *et al.*, 2017; Inoue *et al.*, 2017) and in Kentucky, USA in 2011 (Pratt Katie, 2012). The disease made its first appearance in Bangladesh in 2016, affecting around 15,000 hectares of land and causing a 5-51% reduction in wheat yield in eight southwestern districts including Meherpur, Chuadanga, Kushtia, Pabna, Jessore, Jhenaidah, Bhola, and Barisal (Islam *et al.*, 2016). It was also detected in India in 2017 (Das, 2017) and later in 2020 it was identified in Zambia (Tembo *et al.*, 2020).



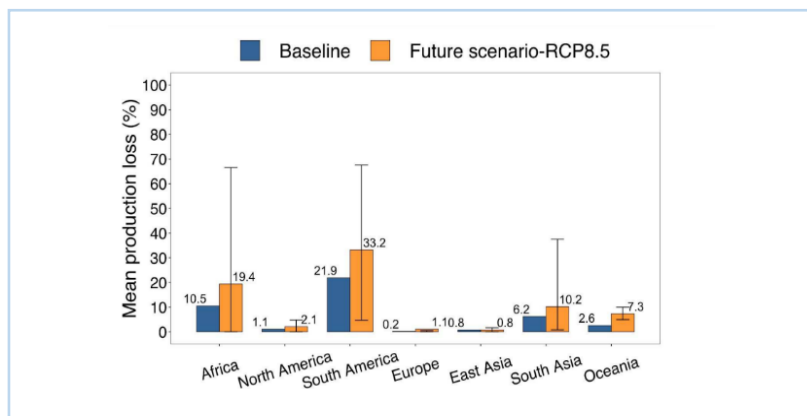
**Figure 1. Global and local spreading of wheat blast disease.** (a) The spreading of wheat blast disease across continents from 1985 to present. (b) The local spreading across Bangladesh from 2016 to present. (Singh *et al.*, 2021).

The occurrence and severity of head blast are influenced by various factors such as environmental conditions, cultivar susceptibility, and plant organ infection (Goulart *et al.*, 2007). Several environmental factors, including higher temperature, rainfall during the flowering stage, leaf/spike wetness, and heavy dew fall, contribute to the outbreak of the disease (Goulart *et al.*, 2007). The outbreak of the wheat blast disease is most severe when there are continuous rainfall and an average temperature of 18-25°C in the flowering stage, followed by warm and humid weather (Kohli *et al.*, 2011). A temperature rise coupled with

rainfall at the flowering time likely contributed to the development of epidemics in the wheat blast-affected districts in Bangladesh in 2016 (Islam *et al.*, 2020).

### 3.1.2 Potential Global Impact of Wheat Blast

In recent years, the world has witnessed several devastating wheat blast epidemics, including the 2016 outbreak in Bangladesh. The underlying cause of these outbreaks is believed to be the increase in temperatures brought about by climate change. Unfortunately, this trend is expected to continue, and new countries are likely to experience similar outbreaks in the future. A crop-disease simulation study revealed that the global risk associated with the spread of wheat blast is considerable. Wheat blast already threatens 6.4 million hectares of crops globally, set to rise to 13 million hectares by mid-century. It could reduce global wheat production by 13%, with South America most affected, followed by African and South Asian countries (Pequeno *et al.*, 2022). These regions are already vulnerable to climate change impacts and have growing wheat consumption. Urgent measures are required to address the problem, including climate change mitigation strategies, resilient crop varieties, and improved agricultural practices to minimize disease outbreaks.



**Figure 2. Simulated vulnerable continental wheat area for wheat blast damage in 1980-2010 baseline and 2040-2070 under climate change. (Pequeno *et al.*, 2022).**

### 3.1.3 Symptoms

Wheat blast is a devastating fungal disease caused by a filamentous fungus, *Magnaporthe oryzae Triticum* (MoT) lineage (synonym *Pyricularia oryzae Triticum* lineage) that can cause up to 100 percent of yield loss. The wheat blast fungus infects all above-ground parts of wheat,

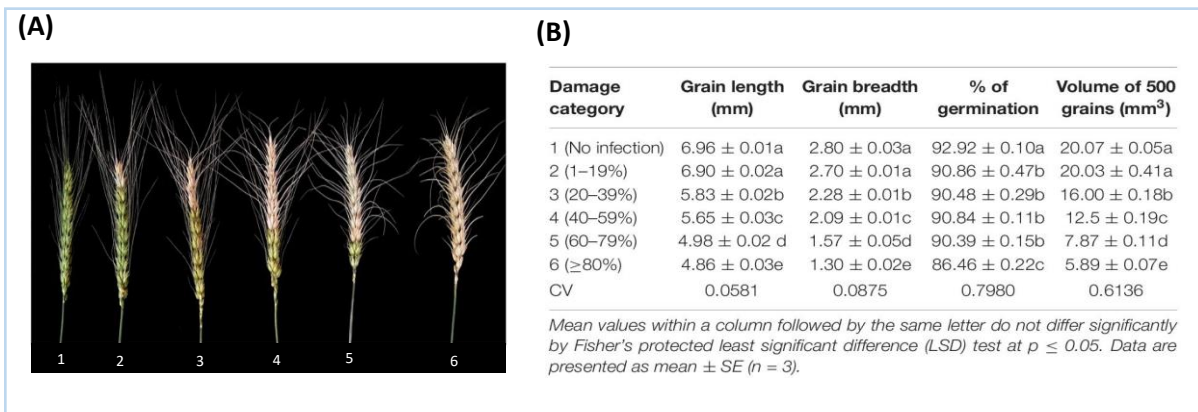
causing partial or complete bleaching on the spikes. Initial symptoms include black spots at the rachis base. Leaves exhibit gray to tan necrotic lesions with dark borders, often with other foliar diseases like spot blotch (Islam *et al.*, 2016). Infection before anthesis or during early flowering leads to sterile spikes and seed abortion. Head infections during flowering result in no grain, while grain filling infections cause small, shriveled, discolored grains (Islam *et al.*, 2016; Urashima *et al.*, 2009).



**Figure 3. Symptoms of wheat blast disease in different plant parts.** (a) complete to partial spike infections. (b) early field infection indicated by a characteristic patch. (c) complete bleaching of 100% spikes. (d) normal color of unaffected wheat grains. (e) severely shriveled, wrinkled, and discolored wheat grains. (f) normal and shriveled grains in a spike. (g) eye-shaped lesion on leaf with gray center. (h) brown lesions with gray centers on an infected wheat stem. (Islam *et al.*, 2020).

#### 3.1.4 Panicle blast impact on grains

MoT infestation had a significant impact on the physical properties of the wheat grain. As the severity of infestation increased, there was a notable decrease in grain length, breadth, volume, and weight per 1000 grains (Surovy *et al.*, 2020).



**Figure 4. Impacts of panicle blast damage severity on wheat grains.** (A) BARI Gom-24 wheat panicles exhibiting varying levels of panicle blast severity (from left to right), category of damage 1–6. (B) Physical changes and germination percentage of wheat grains affected by *Magnaporthe oryzae Triticum* (MoT) infestation. (Surovy *et al.*, 2020).

### 3.1.5 Host specificity

*M. oryzae* is a pathogen that infects different types of grasses. However, there is genetic variation among the pathogen populations that allows them to infect specific host groups. As a result, *M. oryzae* has been divided into different pathotypes based on its ability to infect certain host species (Gladieux *et al.*, 2018). For example, the Triticum pathotype infects wheat, triticale, barley, and several other grass species (Castroagudín *et al.*, 2016; Urashima *et al.*, 1993, 2004). The Avena pathotype infects oats, and the Lolium pathotype infects perennial ryegrass (Farman *et al.*, 2017; Oh *et al.*, 2002).

**Table 1. Various pathotypes of *Magnaporthe oryzae***

Pathotype	Host	Ref
<i>Avena</i> (MoA)	Oats ( <i>Avena sativa</i> ) Wheat ( <i>Triticum aestivum</i> )	(Oh <i>et al.</i> , 2002)
<i>Digitaria</i> (MoD)	Crabgrass ( <i>Digitaria sanguinalis</i> )	(Kato <i>et al.</i> , 2000)
<i>Eleusine</i> (MoE)	Finger millet ( <i>Eleusine coracana</i> )	(Kato <i>et al.</i> , 2000)
<i>Lolium</i> (MoL)	Perennial ryegrass, Wheat ( <i>Triticum aestivum</i> )	(Farman <i>et al.</i> , 2017; Oh <i>et al.</i> , 2002)
<i>Oryzae</i> (MoO)	Rice ( <i>Oryzae sativa</i> )	(Kato <i>et al.</i> , 2000)
<i>Panicum</i> (MoP)	Common millet ( <i>Panicum miliaceum</i> )	(Kato <i>et al.</i> , 2000)

**Table 1. Various pathotypes of *Magnaporthe oryzae* (Cont.)**

<b>Pathotype</b>	<b>Host</b>	<b>Ref</b>
<i>Setaria</i> (MoS)	Foxtail millet ( <i>Setaria italica</i> )	(Kato <i>et al.</i> , 2000)
<i>Triticum</i> (MoT)	Wheat ( <i>Triticum aestivum</i> ), Durum wheat, Barley ( <i>Hordeum sativum</i> ), Rye ( <i>Secale cereale</i> ), Signalgrass ( <i>Urochloa brizantha</i> ) and more than 10 other grass species	(Castroagudín <i>et al.</i> , 2016; Urashima <i>et al.</i> , 1993, 2004)

The evolutionary biology related to the development of the wheat blast fungus pathotype MoT through a host jump has been revealed. Single gene-regulated host specificity has been described in *M. oryzae*, such as the *PWL1* and *PWL2* genes preventing the strains from infecting weeping lovegrass. Five avirulence (AVR) effector genes (*PWT1–5*) have been identified in the population of rice, oat, and *Setaria* pathogens that independently block infection of wheat. Loss of the *PWT3/Rwt3* gene interaction may have contributed to the first emergence of wheat blast in Brazil. Additional circumstantial evidence suggests that wheat varieties planted before the 1980s contained the R gene *Rwt3*, which may have blocked *Lolium* and oat isolates with the corresponding *PWT3* gene from infecting wheat. However, in the 1980s, new *rwt3* wheat varieties lacking this R gene were planted in a new region of Brazil where weather was conducive to blast, allowing the emergence of a *Triticum* population capable of causing disease in all wheat varieties. Nevertheless, all differences in host species specificity are not likely due to AVR-like genes. Further studies are needed to precisely elucidate the underlying molecular mechanisms of host-specificity and host range of *M. oryzae* pathotypes, and factors involved with the host jumps.

## **3.2 Genomic resources for wheat blast**

### **3.2.1 QTL mapping for wheat blast resistance**

QTL (Quantitative Trait Locus) mapping is a type of genetic mapping that helps to identify the specific regions on a chromosome where these genes are located and estimate their contribution to the trait of interest. Genetic mapping has helped breeders in improving various plant species by identifying economically important genes responsible for complex traits like disease resistance (Ashkani *et al.*, 2016). This process has been successful in rice, where more than

350 QTLs (Quantitative Trait Locus) for disease resistance have been identified (Ashkani *et al.*, 2016). However, the research on QTL mapping for resistance to wheat blast disease is limited, and only a few major and stable QTLs have been identified (Table 2) beyond the 2AS/2NS translocation (Singh *et al.*, 2021b).

**Table 2. Identified QTLs for wheat blast resistance to date** (Nizolli *et al.*, 2023)

QTL number	DNA markers	Mapping type	Ref.
QWbr.emt-2 <sup>a</sup>	KASP and SSRs	Linkage mapping	(Ferreira <i>et al.</i> , 2021)
QPag.emt-2 <sup>a</sup>	KASP and SSRs	Linkage mapping	(Ferreira <i>et al.</i> , 2021)
QWbr.emt-5B	KASP and SSRs	Linkage mapping	(Ferreira <i>et al.</i> , 2021)
QWbr.emt-7B	KASP and SSRs	Linkage mapping	(Ferreira <i>et al.</i> , 2021)
Loco 2AS	DArTSeq and STS	Linkage mapping	(He <i>et al.</i> , 2021)
Loco 2DL	DArTSeq and STS	Linkage mapping	(He <i>et al.</i> , 2021)
Loco 7AL	DArTSeq and STS	Linkage mapping	(He <i>et al.</i> , 2021)
Loco 7DS	DArTSeq and STS	Linkage mapping	(He <i>et al.</i> , 2021)
Loco 2AS	SNP	Association mapping	(Juliana <i>et al.</i> , 2020)
Loco 3BL	SNP	Association mapping	(Juliana <i>et al.</i> , 2020)
Loco 4AL	SNP	Association mapping	(Juliana <i>et al.</i> , 2020)
Loco 7BL	SNP	Association mapping	(Juliana <i>et al.</i> , 2020)
Loco 1AS	STS	Association mapping	(He <i>et al.</i> , 2020)
Loco 2BL	STS	Association mapping	(He <i>et al.</i> , 2020)
Loco 3AL	STS	Association mapping	(He <i>et al.</i> , 2020)
Loco 4BS	STS	Association mapping	(He <i>et al.</i> , 2020)
Loco 4DL	STS	Association mapping	(He <i>et al.</i> , 2020)
Loco 7BS	STS	Association mapping	(He <i>et al.</i> , 2020)
Loco 2A	SNP	Association mapping	(Cruppe <i>et al.</i> , 2021)
Loco 1BS	SNP and STS	Association mapping	(He <i>et al.</i> , 2021)
Loco 2AS	SNP and STS	Association mapping	(He <i>et al.</i> , 2021)
Loco 6BS	SNP and STS	Association mapping	(He <i>et al.</i> , 2021)
Loco 7BL	SNP and STS	Association mapping	(He <i>et al.</i> , 2021)
Loco 1A	SNP	Association mapping	(Goddard <i>et al.</i> , 2020)
Loco 2B	SNP	Association mapping	(Goddard <i>et al.</i> , 2020)
Loco 4A	SNP	Association mapping	(Goddard <i>et al.</i> , 2020)
Loco 5A	SNP	Association mapping	(Goddard <i>et al.</i> , 2020)

### 3.2.2 Status of resistant genes to date

Several genes have already been identified that confer resistance against blast disease (Table 3).

**Table 3. Identified blast-resistant genes to date** (Singh *et al.*, 2021b)

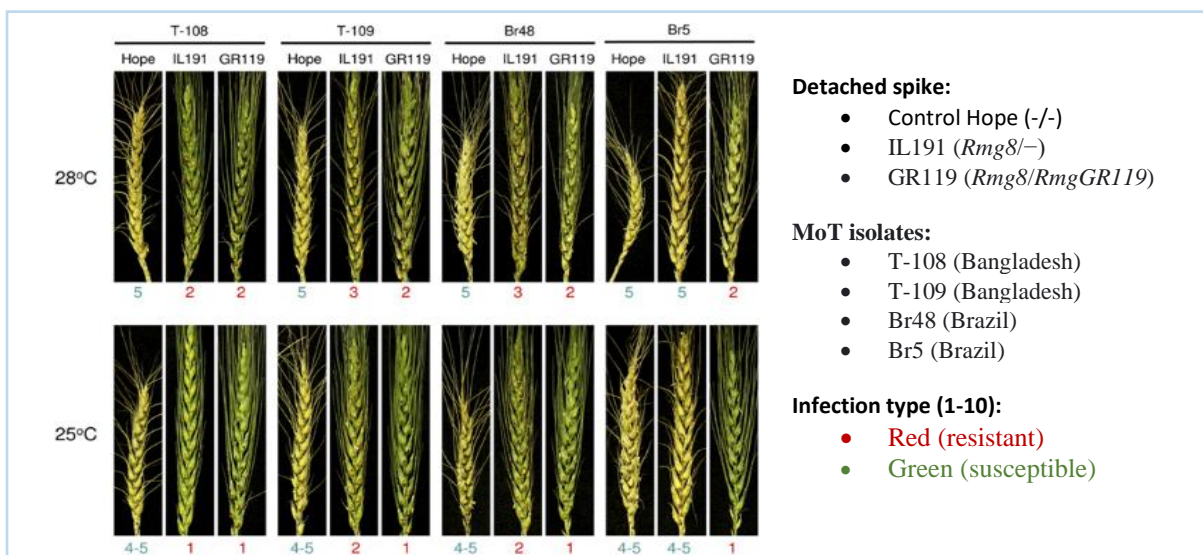
R gene	Source wheat species	Cultivar	Chromosome Location	Wheat blast strain	Efficacy of the gene	Ref.
<b><i>RmgTd (t)</i></b>	<i>Triticum dicoccum</i>	Ku109	-	A mutant progeny of MoA and MoT	Confer moderate resistance	(Cumagun <i>et al.</i> , 2014)
<b><i>Rmg1</i></b>	<i>Triticum aestivum</i>	Norin 4	1D	MoA	Confer resistance in seedlings and heads, but temperature sensitive	(Takabayashi <i>et al.</i> , 2002)
<b><i>Rmg2</i></b>	<i>Triticum aestivum</i>	Thatcher	7A	MoT	Confer resistance at seedling stage, temperature sensitive	(Zhan <i>et al.</i> , 2008)
<b><i>Rmg3</i></b>	<i>Triticum aestivum</i>	Thatcher	6B	MoD	Confer high resistance at a high temperature (26°C)	(Zhan <i>et al.</i> , 2008)
<b><i>Rmg4</i></b>	<i>Triticum aestivum</i>	Norin 4	4A	MoD	Confer high resistance at a high temperature (26°C)	(Nga <i>et al.</i> , 2009)

**Table 3. Identified blast-resistant genes to date (Cont.)**

<b>R gene</b>	<b>Source wheat species</b>	<b>Cultivar</b>	<b>Chromosome Location</b>	<b>Wheat blast strain</b>	<b>Efficacy of the gene</b>	<b>Ref.</b>
<b><i>Rmg5</i></b>	<i>Triticum aestivum</i>	Red Egyptian	6D	MoD	Confer resistance in seedlings and heads, but temperature-sensitive	(Nga <i>et al.</i> , 2009)
<b><i>Rmg6</i></b>	<i>Triticum aestivum</i>	Norin 4	1D	MoL, MoE, MoA	Confer resistance at the heading stage but ineffective at 26°C	(Vy <i>et al.</i> , 2014)
<b><i>Rmg7</i></b>	<i>Triticum dicoccum</i>	St24, ST17, ST25	2A	MoT	Confer resistance at the heading stage but ineffective at 26°C	(Tagle <i>et al.</i> , 2015)
<b><i>Rmg8</i></b>	<i>Triticum aestivum</i>	S-615	2B	MoT	Confer resistance at the heading stage and even at 26°C	(Anh <i>et al.</i> , 2015)
<b><i>2NS/2A S</i></b>	<i>Aegilops ventricos a</i>	-	-	MoT	Confer resistance to head blast, but not foliar blast	(Cruz <i>et al.</i> , 2016)
<b><i>RmgG R119</i></b>	Albanian wheat	GR119	-	MoT	Confer high resistance to all Triticum isolates tested	(Wang <i>et al.</i> , 2018)



However, genes including *Rmg2*, *Rmg3*, and *Rmg7* are ineffective at the head stage, and the resistance conferred by 2NS translocation was eroded by a highly aggressive new isolate, B-71, in some wheat genetic backgrounds (Cruppe *et al.*, 2020). Only *Rmg8* and *RmgGR119* demonstrated resistance to wheat blast at both the seedling and head stages and are not temperature labile (Tagle *et al.*, 2015; Anh *et al.*, 2018, Latorre *et al.*, 2023)). *RmgGR119* was reported to be effective against a variety of MoT isolates in the Albanian wheat landrace GR119. This landrace also possesses *Rmg8*, implying that *Rmg8* and *RmgGR119* gave a high level of blast resistance when combined (Wang *et al.*, 2018). However, their performance on head blast must be evaluated in the field before they can be used in a breeding program.



**Figure 5.** Reactions of detached wheat spikes of Hope (-/-), IL191 (*Rmg8*/-), and GR119 (*Rmg8*/*RmgGR119*) to MoT isolates. (Horo *et al.*, 2020).

### 3.2.3 Status of current resistant varieties

Subsequent screening studies in Brazil, Bolivia, and Paraguay led to the identification of a few moderately resistant varieties (Table 4). Many of these varieties have the CIMMYT genotype Milan in their pedigree, and it was later discovered that the 2NS translocation was the underlying resistance factor (Cruz *et al.*, 2016). 2NS was first transferred from *Aegilops ventricosa* to a French variety "VPM1" (Helguera *et al.*, 2003). This translocation confers resistance to a wide range of plant diseases and also increases yield potential. However, the translocation has different effects on wheat lines, indicating the role of genetic background in its expression. "BARI Gom-33," a zinc bio-fortified blast-resistant wheat variety released in Bangladesh in 2017, is a success story of utilizing the 2AS/2NS translocation in breeding

(Hossain *et al.*, 2019). However, over-reliance on this translocation in South Asia and South America makes it vulnerable to new MoT isolates due to strong directional selection (Cruz *et al.*, 2016). Moderately resistant non-2NS sources and wild wheat relatives (*Ae. tauschii* and *Ae. Umbellulata*) have also been identified as potential sources of resistance against WB (He *et al.*, 2021). However, several resistant sources must be thoroughly tested to ensure their effectiveness under various environmental conditions.

**Table 4. Commercialized blast-resistant varieties to date**

Variety	Country	Resistance level	Background	Ref.
BARI Gom 33	Bangladesh	High	2NS	(Hossain <i>et al.</i> , 2019)
Borloug 100	Bangladesh, Bolivia, Nepal	High	2NS	(Singh <i>et al.</i> , 2021a)
BR 18-Terena	Brazil	High	Non 2NS	(Goddard <i>et al.</i> , 2020)
BR8	Brazil	High	2AS/2NS	(Ha <i>et al.</i> , 2016)
BRS 229	Brazil	High	Non 2NS	(Brunetta <i>et al.</i> , 2006)
Caninde 1“S”	Paraguay	High	2AS/2NS	(Ha <i>et al.</i> , 2016)
Milan	South America	High	2AS/2NS	(Ha <i>et al.</i> , 2016)
Paragua CIAT	Bolivia	High	-	(Kohli <i>et al.</i> , 2011)
Parapeti CIAT	Bolivia	High	-	(Kohli <i>et al.</i> , 2011)
BRS 120	Brazil	Moderate	2NS	(Prestes <i>et al.</i> , 2007)
BRS 220	Brazil	Moderate	2NS	(Prestes <i>et al.</i> , 2007)
BRS 49	Brazil	Moderate	2NS	(Prestes <i>et al.</i> , 2007)
Caninde 1	Paraguay	Moderate	2NS	(Kohli <i>et al.</i> , 2011)
CD 116	Brazil	Moderate	2NS	(Prestes <i>et al.</i> , 2007)
IAPAR 53	Brazil	Moderate	-	(Prestes <i>et al.</i> , 2007)
IPR 85	Brazil	Moderate	-	(Kohli <i>et al.</i> , 2011)
Itapua 75	Paraguay	Moderate	2NS	(Buerstmayr <i>et al.</i> , 2017)
Motacu CIAT	Bolivia	Moderate	Non 2NS	(Buerstmayr <i>et al.</i> , 2017)
Patuju CIAT	Bolivia	Moderate	Non 2NS	(Buerstmayr <i>et al.</i> , 2017)
Sausal CIAT	Bolivia	Moderate	2AS/2NS	(Buerstmayr <i>et al.</i> , 2017)

### 3.3 Deployment of the current knowledge for durable blast resistance of wheat

Two strategies for developing durable and robust blast-resistant wheat varieties are screening for more resistant genes and incorporating them into local cultivars, and using gene editing techniques to silence genes that make plants susceptible to blast disease (Islam *et al.*, 2022). Utilizing bio fungicide can also be a potential novel strategy in mitigating wheat blast disease.

#### 3.3.1 Introgression of resistant genes into local cultivar

Some of the wheat blast-resistant genes have already been integrated into elite wheat varieties by Japanese scientists (Wang *et al.*, 2018). *Rmg8* and *RmgGR119* are currently the two bona fide blast-resistant genes as the 2NS translocation has been eroded by a new, highly aggressive isolate called B-71 in certain genetic backgrounds, as stated by (Cruz *et al.*, 2016). To confer durable blast resistance, scientists recommend the incorporation of multiple resistant genes in a single cultivar. Therefore, researchers at the Institute of Biotechnology and Genetic Engineering (IBGE) of Bangabandhu Sheikh Mujibur Rahman Agricultural University in Bangladesh are utilizing marker-assisted selection (MAS) to introduce *Rmg8* and *RmgGR119* genes into a biofortified, zinc-enriched variety called BARI Gom-33, which already possesses the 2NS translocation (Singh *et al.*, 2021b).

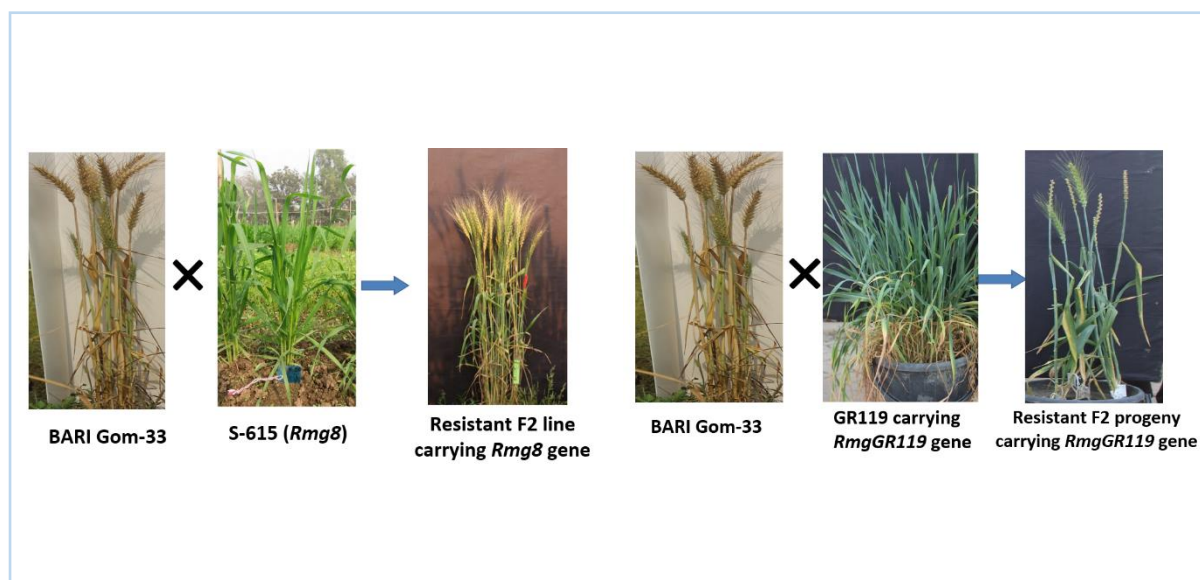


Figure 6. Introgression of *Rmg8* and *RmgGR119* genes into BARI Gom-33

#### 3.3.2 Mutation breeding potential

The IAEA database reports that various countries have employed mutation breeding programs that have resulted in the creation of 69 wheat cultivars with resistance to different fungal infections (Singh *et al.*, 2021b). In 2018, a mutant wheat variety was released in Mongolia

which was resistant to spots, loose smut, and corrosion of stripes, *Septoria nodorum* blotch (SNB), and *Alternaria* leaf blight (IAEA, 2018). Additionally, mutation breeding has been successful in producing wheat varieties resistant to Ug99 in Kenya, with Eldo Ngano1 and Eldo Mavuno1 being two cultivars that were made available to Kenyan farmers in 2014 (Bado, 2015).

In 2019, a mutation breeding approach was applied in Bangladesh to develop blast-resistant wheat. Researchers led by Harun-Or-Rashid conducted a screening of the M2 population of gamma-irradiated wheat, with four different radiation doses (150 GY, 200 GY, 250 GY, and 300 GY) being applied to the seeds of three wheat varieties: BARI Gom-25, BARI Gom-29, and BARI Gom-30.

**Table 5. Incidence and severity of wheat blast on gamma irradiation treated seeds**

(Harun-Or-Rashid *et al.*, 2019)

Irradiation	BARI Gom-25		BARI Gom-29		BARI Gom-30	
	Disease	Disease	Disease	Disease	Disease	Disease
	Incidence	Severity	Incidence	Severity	Incidence	Severity
	(%)	(%)	(%)	(%)	(%)	(%)
No	56.66 a	87.33 a	48.00 a	94.00 a	34.33 a	85.00 a
Irradiation						
150 GY	37.33 c	80.00 b	30.00 c	81.66 b	29.33 b	74.33 b
200 GY	22.33 c	42.33 d	28.66 c	72.66 b	18.00 c	51.66 e
250 GY	32.33 c	52.33 c	17.33 d	60.00 c	25.00 b	57.66 e
300 GY	44.00 b	75.00 b	36.00 b	77.66 b	28.66 b	68.33 c

The letters denote statistical differences at the 95% level using Turkey’s HSD (Honestly significant difference) test.

The results of the screening showed that the radiation doses of 200 GY and 250 GY were more effective in reducing the incidence and severity of wheat blast. Among the three wheat varieties evaluated, BARI Gom-30 demonstrated the best performance, followed by BARI Gom-25 and BARI Gom-29. The team recommends advancing these mutant lines to the M3 to M5 stages and artificially inoculating them with the wheat blast pathogen to evaluate their level of resistance (Harun-Or-Rashid *et al.*, 2019). The number of currently known “R” genes for wheat blast is low. Hence, mutations can help in identifying novel “R” genes along with modifying the existing ones for improving wheat blast resistance (Singh *et al.*, 2021b).

### 3.3.3 Genome editing approaches

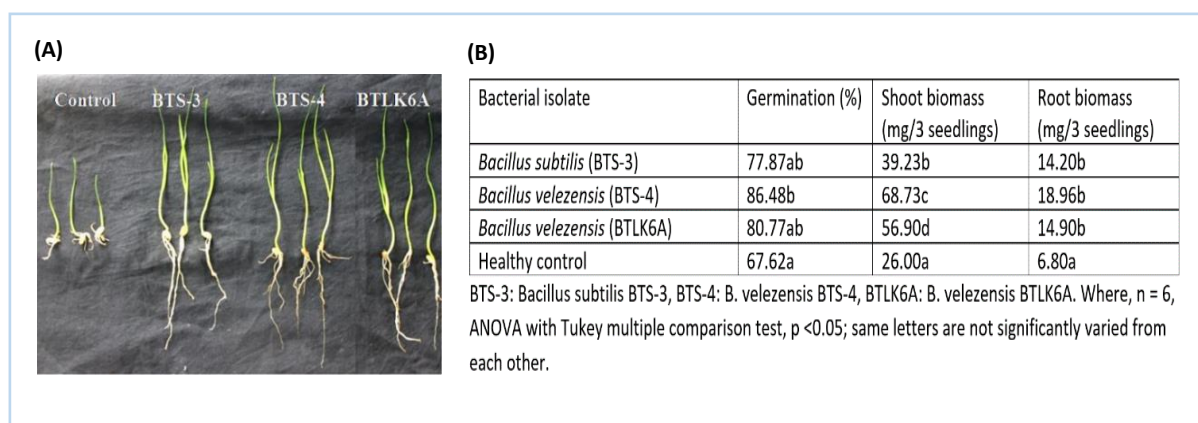
CRISPR-Cas9 genome editing toolkits have great potential in developing durable blast-resistant wheat varieties (Islam, 2019a; Islam *et al.*, 2019b, 2020) and have already been used to develop blast-resistant mutagenized rice lines by knocking out the rice gene *OsSEC3A* (S gene responsible for rice blast) (Ma *et al.*, 2018), while in wheat, it has demonstrated its potential to mutate wheat blast susceptibility genes by disrupting genes like *TaDREB2* and *TaERF3* (Kim *et al.*, 2018). Introducing R genes is a promising method to confer wheat blast resistance, but the resistant genes have not been yet cloned and characterized. Again, wheat blast is a new disease, and there is no reported S gene in wheat. Scientists have developed a robust, transgene-free, tissue culture-free approach for genome editing through virus infection (Li *et al.*, 2021), and also developed a heritable editing procedure in planta through grafting (Yang *et al.*, 2023). Therefore, cloning and characterization of R genes, and identification of S genes, are required for a successful genome editing approach to combat wheat blast (Islam *et al.*, 2022).

**Table 6. CRISPR/Cas9-mediated resistance against plant pathogens**

Plant species	Targeted genes	Disease/Pathogen	Ref.
<i>Arabidopsis thaliana</i>	<i>eIF(iso)4E</i>	Turnip mosaic virus (TuMV)	(Pyott <i>et al.</i> , 2016)
Citrus	<i>CsLOB1</i>	Citrus canker ( <i>Xanthomonas citri</i> subsp. <i>Citri</i> )	(Peng <i>et al.</i> , 2017)
Cucumber	<i>eIF4E</i>	Papaya ring spot mosaic virus (PRSMV) Zucchini yellow mosaic virus (ZYMV) Cucumber Vein Yellowing Virus (CVYV)	(Chandrasekaran <i>et al.</i> , 2016)
<i>Nicotiana benthamiana</i>	Coat proteins	Tomato yellow leaf curl virus (TYLCV) Beet curly top virus (BCTV)	(Ali <i>et al.</i> , 2016)
Rice	<i>OsERF922</i>	Rice blast ( <i>Magnaporthe oryzae</i> <i>Oryzae</i> )	(Wang <i>et al.</i> , 2016)
Tomato	<i>SIMlo</i>	Powdery mildew ( <i>Podosphaera xanthii</i> )	(Nekrasov <i>et al.</i> , 2017)
Wheat	<i>TaMlo</i>	Powdery mildew ( <i>Blumeria graminis</i> f. sp. <i>Tritici</i> )	(Wang <i>et al.</i> , 2018)

### 3.3.4 Biological control

Biological control of plant diseases is becoming increasingly popular as it offers several advantages over chemical control methods such as being eco-friendly, cost-effective, and sustainable (Mia *et al.*, 2023). After screening 170 bacterial isolates, three strains of *Bacillus* species (BTS-3, BTS-4, and BTLK6A) were identified as potential antagonists reducing 89, 88, and 85% of wheat blast disease severity, respectively, compared to mock-inoculated control against MoT in vitro (Surovy *et al.*, 2022).



**Figure 7. Antagonistic effect of three probiotic bacterial strains on blast infected seeds.**

(A) Enhancement of wheat seedling growth by seed endophytic probiotic bacteria. (B) Enhancement of seed germination, and shoot-root biomass of wheat seedling cv. BARI Gom 24 (Prodip) by using seed endophytic probiotic bacteria. (Surovy *et al.*, 2022)

**Table 7. Antagonistic effects of bioactive natural products on MoT**

Bioactive compound	Activity	Mode of action	Source	Conc. (µg/ml)	Blast inhibition	Ref.	
Antimycin A	Mycelial asexual life phases of MoT, conidiogenesis, and MoT suppression	growth,	-	<i>Streptomyces</i> sp	10	In vitro and in vivo	(Paul <i>et al.</i> , 2022)

**Table 7. Antagonistic effects of bioactive natural products on MoT (Cont.)**

<b>Bioactive compound</b>	<b>Activity</b>	<b>Mode of action</b>	<b>Source</b>	<b>Conc. (µg/ml)</b>	<b>Blast inhibition</b>	<b>Ref.</b>
Bonactin	Mycelial growth, asexual life phases of MoT, and MoT suppression	-	<i>Streptomyces</i> spp.	5	In vivo and in vitro	(Rabby <i>et al.</i> , 2022)
Chelerythrine	Mycelial growth, asexual life phases of MoT, conidiogenesis, and MoT suppression	Inhibits protein kinase activity	<i>Chelidonium majus</i> , <i>Macleaya cordata</i>	50	In vitro and in vivo	(Chakra borty <i>et al.</i> , 2022)
Feigrisolide C	Mycelial growth, asexual life phases of MoT, conidiogenesis, and MoT suppression	-	<i>Streptomyces</i> spp.	5	In vivo and in vitro	(Rabby <i>et al.</i> , 2022)
Linear lipopeptides	Mycelial growth, conidiogenesis, conidial germination inhibition, and MoT suppression	-	<i>Bacillus subtilis</i>	10	In vivo	(Chakra borty <i>et al.</i> , 2020a)
Oligomycin B	Mycelial growth, conidiogenesis, conidial germination inhibition, and MoT suppression	Mitochondrial ATPase inhibition	<i>Streptomyces</i> sp.	10	In vivo	(Chakra borty <i>et al.</i> , 2020)
Oligomycin F	Mycelial growth, conidiogenesis, conidial germination inhibition, and MoT suppression	Mitochondrial ATPase inhibition	<i>Streptomyces</i> sp.	10	In vivo	(Chakra borty <i>et al.</i> , 2020b)
Staurosporine	Mycelial growth, asexual life phases of MoT, and MoT suppression	Inhibits protein kinase activity	<i>S. staurosporeus</i> <i>S. roseoflavus</i> .	50	In vitro and in vivo	(Chakra borty <i>et al.</i> , 2022)

Researchers have screened several bioactive natural products against MoT and discovered that secondary metabolites from microorganisms and plants, such as lipopeptides, macrolides, alkaloids, terpenoids, and phenolics, effectively suppress MoT growth (Table 7). To determine the effectiveness of these secondary metabolites as fungicides against wheat blast disease, a field evaluation is required, which could pave the way for the development of a new biofungicide (Chakraborty *et al.*, 2022; Chakraborty *et al.*, 2020; Paul *et al.*, 2022; Rabby *et al.*, 2022) e.



## CHAPTER IV

### CONCLUSIONS

This wheat blast disease has the potential to drastically lower crop production and jeopardize food and economic security in areas where it has spread. Wheat blast's potential to inflict extensive losses necessitates a quick effort to better understand and manage this deadly disease. Unlike rice blast and MoO pathosystem, the mechanisms that govern the interaction between the wheat plant and MoT and determine disease development are not yet fully studied. More research is needed to investigate the molecular factors behind host-specificity and host-range of *M. oryzae* pathotypes, as well as the factors that contribute to host-jumps.

Additional screening for host resistance genes and QTLs is required, alongside the integration of *Rmg8* and *RmgGR119* genes into local high yielding cultivars, followed by field assessment. Head blast evaluation should be prioritized over seedling infection. Combining multiple resistant genes in a single cultivar can provide durable blast resistance.

Identification of more R genes as well as S genes in wheat followed by cloning and characterization of them is crucial for successful genome editing approach for developing panicle blast resistance. CRISPR/Cas can be used as it now offers a heritable, transgene free edits in planta. Additionally, field evaluation and characterization of antimicrobial compounds are necessary for a better understanding of their mode of action and practical recommendations for wheat blast control in farmers' fields.

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