Plant Archives Vol. 25, Supplement 2, 2025 pp.1221-1228 e-ISSN:2581-6063 (online), ISSN:0972-5210

Plant Archives

Journal homepage: http://www.plantarchives.org
DOI Url : https://doi.org/10.51470/PLANTARCHIVES.2025.v25.supplement-2.152

STUDY OF TEMPORAL DYNAMICS OF *MAGNAPORTHE ORYZAE* VIRULENCE IN RICE AND RACE STRUCTURE USING DIFFERENTIAL CULTIVARS AND MONOGENIC LINES

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(Date of Receiving : 01-04-2025; Date of Acceptance : 09-06-2025)

ABSTRACT

Rice blast, caused by *Magnaporthe oryzae*, poses a persistent threat to global rice production due to its high virulence diversity and adaptability. This study aimed to monitor the race structure of *M. oryzae* populations using an international set of eight differential cultivars and a comprehensive panel of monogenic lines carrying known blast resistance (R) genes. Field and nursery experiments were conducted over two growing seasons (2023 and 2024) at different growth stages of rice. Disease scoring and race designation were performed using standard international protocols, and the corresponding avirulence (*Avr*) gene presence in the pathogen was inferred based on observed incompatibility reactions. Results revealed the emergence of multiple races across both years and developmental stages, highlighting the temporal variability in virulence structure. Several R genes, including *Pi1, Pi2, Pi54, Pi9, Pi-7*, and *Pi-12*, conferred high levels of resistance. Moderate resistance in some lines suggested the presence of slow-blasting resistance mechanisms. The findings underscore the importance of continuous virulence monitoring to guide the strategic deployment of R genes and support durable blast resistance breeding programs.
Keywords : Rice blast, *Magnaporthe oryzae*, virulence monitoring, differential cultivars, monogenic lines

Introduction

Rice blast, caused by the fungal pathogen *Magnaporthe oryzae*, is one of the most destructive diseases affecting rice production globally. The disease leads to significant yield losses by infecting various parts of the rice plant, including leaves, nodes, and panicles (Talbot, 2003). Understanding and monitoring the virulence dynamics of *M. oryzae* populations are essential for managing this pathogen effectively and deploying resistant cultivars strategically.

The rice *M. oryzae* pathosystem is governed by a classical gene-for-gene relationship, where resistance (R) genes in the host recognize specific avirulence (*Avr*) genes in the pathogen (Flor, 1971). This recognition triggers a defense response, often culminating in localized cell death to prevent further pathogen spread. However, the widespread cultivation of rice varieties with single R genes imposes strong selection pressure on pathogen populations, resulting in the emergence of virulent races that overcome host resistance through mutations, deletions, or insertions in *Avr* genes (Valent & Khang, 2010). As of now, over 118 R genes and QTLs conferring resistance to blast have been identified, with 36 cloned and functionally characterized (Ashkani *et al*., 2015). Similarly, around 40 *Avr* genes have been genetically analyzed, with many cloned, offering valuable insights into host-pathogen interactions. Monitoring the race structure of *M. oryzae* across different temporal and spatial scales helps in understanding the adaptability of the pathogen and guides resistance breeding programs. Sets of differential cultivars are an effective means of tracking the dynamics of pathogen populations in several host–

pathogen systems (Atkins *et al*., 1967; Feng *et al*., 2018; Fernando *et al*., 2018; Liu *et al*., 2017; Wang *et al*., 2017; Zhang *et al*., 2017, 2019). Because an understanding of the race structure of a pathogen population supports a rational deployment of host resistance cultivars, the literature includes several studies directed to this goal.

The present investigation utilized both an international set of differential rice cultivars and a comprehensive collection of monogenic lines to monitor the virulence patterns of *M. oryzae* populations over two consecutive years. The study aimed to identify prevalent races, speculate corresponding *Avr* gene presence, and evaluate the effectiveness of known R genes across different developmental stages of the rice crop. Such studies are crucial in informing rice breeding strategies and in deploying durable resistance genes to manage blast disease effectively.

Materials and Methods

Materials

Population for virulence monitoring of blast

The international set of eight differential cultivars A=**Raminad -STR -3;** B=**Zenith;** C=**NP – 125;** D=**USEN;** E=**Dular;** F=**Kanto – 51;** G=**Shi-tia-tao;** H=**Calaro** proposed by United states-Japan cooperative blast project was used. These differentials each carry distinct blast resistance (R) genes, enabling the assessment of pathogen virulence based on specific host-pathogen interactions. (Table-1).

Table 1 : Set of 8 differential cultivar used for virulence monitoring

Letters assigned	Cultivar	Blast Resistance Gene(s)
A	Raminad Strain 3	-
B	Zenith	*Pi-z + Pi-a + Pi-i*
C	NP125	*Pi-k*c*
D	USEN	*Pi-a+(=Pi4(t))*
E	Dular	*Pi-ka+*
F	Kanto-51	*Pi-k*
G	Shi Tia Tao	*Pi-ks*
H	Calaro	*Pi-9*

In addition to the international differential set, the study employed a comprehensive collection of monogenic lines and genotypes specifically developed for blast resistance research. These included RP Patho and RP Biopatho lines, recombinant inbred lines (RILs), and pre-breeding selections (PRS lines), each representing a distinct genetic background and carrying one or more characterized blast resistance (R) genes. Most of these lines were developed through systematic backcrossing into susceptible indica or japonica backgrounds such as CO39 or Lijiangxintuanheigu (LTH), resulting in near-isogenic lines (NILs) that differ primarily by the resistance gene of interest. This approach ensures that phenotypic differences in disease response can be attributed directly to the presence or absence of specific R genes (Table-2).

Table 2 : International set of 39 monogenic lines used for virulence monitoring

Line no.	Genotype	Gene	Line no.	Genotype	Gene
1	C101 LAC	*Pi1*	26	RP Patho-1	*Pi1*
2	C101 A51	*Pi2*	27	RP Patho-2	*Pi2*
3	C104 AKT	-	28	RP Patho-3	*Pi54*
4	C101 TTP	*Pi-4b*	29	RP Patho-7	*Pi1*
5	RIL - 10	*Pi-12*	30	RP Patho-8	*Pi2*
6	RIL - 29	*Pi-7*	31	RP Patho-9	*Pi54*
7	O. minuta	*Pi9*	32	RP Biopatho-1	*Pi2*
8	BL-122	*Pi-1 + Pi-2*	33	RP Biopatho-2	*Pi54*
9	BL-245	*Pi-2 + Pi-4*	34	RP Biopatho-3	*Pi2*
10	A 57	*Pi-1 + Pi-2 + Pi-4*	35	RP Biopatho-4	*Pi54*
11	C101 PKT	*Pi-3*	36	PRS-17	*Pi9+Pi54*
20	Tadukan	*Pi-ta*	37	PRS-50 (RP 6618)	*Pi54*
21	IR - 64	Resistant	38	RP 6617-58(PRS-58)	*Pi9*
22	Tetep	*Pi-kh+*	39	RP 6617-59(PRS-59)	*Pi9*
23	HR - 12	Susceptible			
24	Rasi	Resistant			
25	Co-39	Susceptible			

Methods

The experimental investigation was carried out over two consecutive years, 2023 and 2024, encompassing two distinct stages of rice growth: the nursery stage and the vegetative (adult) phase. The nursery bed was arranged with two rows of differential lines alternated with a spreader line *i.e.,* Swarna (susceptible check). Stage I blast disease reactions were recorded at the nursery level on 17 August 2023 and 31 August 2024. In the field trials, disease assessments were conducted at 21 (stage II) and 35 (stage III) days after transplanting (DAT) in 2023, and at 19 (stage II) and 38 (stage III) DAT in 2024. The disease reaction of each differential and monogenic line was scored using Standard Evaluation System (SES) for rice developed by IRRI. Pathogen race designation was based on the reaction patterns of the eight international differentials using the international race identification number proposed by K.C. Ling and S.H. Ou, 1968.

Result and Discussion

Designating the races based on the reaction of international rice differentials to *Magnaporthe oryzae* in Jagdalpur

Blast disease assessments scores were used to designate race number for each year and crop stage. *Magnaporthe oryzae* race number was compared with the pre-designated pathogenicity patterns of all races. Since the reaction of differential in our case was intermediate type the lower-case latter (representing respectively the eight-differential variety) after the race number for indicating the intermediate reaction for the respective differential, (*designation for intermediate reaction type* a=Raminad -STR -3; b=Zenith; c=NP – 125; d=USEN; e=Dular; f=Kanto – 51; g=Shi-tia-tao; h=Calaro). This notation provides a more nuanced representation of the pathogen's virulence profile, capturing partial resistance and moderate susceptibility in the host-pathogen interaction.

1. As pre-designated pathogenicity patterns for the surviving *Magnaporthe oryzae* population were IE244^{abcgh}. The *Magnaporthe oryzae* population expressed a resistant reaction with a score 1 on D= USEN while the rest of the 07-differential expressed MR / MS reaction type.
2. Similarly, based on observed blast score on 31st August 2024 (Table-3) the race designation conferred to the surviving *Magnaporthe oryzae* population was II256^{adefgh}.
3. The international set of eight differential expressed during the two consecutive years (at seedling stage in the nursery on 17th August 2023 and on 31st August 2024) varied and revealed the presence of two different races IE244^{abcgh}, II256^{adefgh}.
4. The international set of eight differential cultivars were also used to analyze specific interactions at 21 days (Stage II) and 35 days (Stage III) during 2023 (Table-4) and at 19 days (Stage II) and 38 days (Stage III) expressed during 2024 (Table-5).
5. The rice crop was at the vegetative phase at which scoring of blast disease was done. The rice crop at the vegetative phase constitutes a different physiology than the seedling in the nursery stage. Two races were resolved IG254^{abdfh} (21 days); IB128^{ace} (35 days) during the year 2023; and ID231^{acefh} (19 days); ID240^{cefgh} (38 days) during the year 2024.

Table 3: Expression of eight international rice differentials at Stage I - nursery stage against the *Magnaporthe oryzae* population in Jagdalpur.

Differentials	Name of Differentials	Gene(s)	Stage I			
			Year 2023		Year 2024	
			Score	Reaction	Score	Reaction
A	Raminad -STR -3	*unknown*	2	MR	2	MR
B	Zenith (U.S.)	*Pi-z + Pi-a + Pi-i*	2	MR	1	R
C	NP - 125	*Pik*c*	2	MR	1	R
D	USEN (China)	*Pi-a+(= Pi4(t))*	1	R	3	MR
E	Dular (Pakistan)	*Pi-ka+*	5	MS	2	MR
F	Kanto - 51	*Pi-k*	5	MS	3	MR
G	Shi-tia-tao (China)	*Pi-ks*	3	MR	3	MR
H	Calaro (U.S.)	*Pi-9*	3	MR	2	MR
Designated race number				**IE244^{abcgh}**		**II256^{adefgh}**

For intermediate reaction for A=Raminad -STR-3; B=Zenith; C=NP–125; D=USEN; E=Dular; F=Kanto – 51; G=Shi-tia-tao; H=Calaro lower case alphabets are used for respective differntials a, b, c, d, e, f, g, h as a suffix to the race designation

Table 4: Expression of eight international rice differentials at stage II and III against the *Magnaporthe oryzae* population in Jagdalpur during 2023.

Line no.	Differentials	Gene(s)	Stage II (21 days)		Stage III (35 days)	
			Score	Reaction	Score	Reaction
A	Raminad -STR -3	*unknown*	3.5	MS	5	MS
B	Zenith (U.S.)	*Pi-z + Pi-a + Pi-i*	4	MS	5.5	S
C	NP - 125	*Pik*c*	2	R	4	MS
D	USEN (China)	*Pi-a+(= Pi4(t))*	5	MS	7	S
E	Dular (Pakistan)	*Pi-ka+*	2	R	3.5	MS
F	Kanto - 51	*Pi-k*	4.5	MS	6.5	S
G	Shi-tia-tao (China)	*Pi-ks*	6.5	S	9	HS
H	Calaro (U. S.)	*Pi-9*	4	MS	6.5	S
Designated race number				**IG254^{abdfh}**		**IB128^{ace}**

Table 5: Expression of eight international rice differentials at stage II and III against the *Magnaporthe oryzae* population in Jagdalpur during 2024.

Line no.	Differentials	Gene(s)	Stage II (19 days)		Stage III (38 days)	
			Score	Reaction	Score	Reaction
A	Raminad -STR -3	*unknown*	2.5	MR	1.5	R
B	Zenith (U.S.)	*Pi-z + Pi-a + Pi-i*	0.5	R	1	R
C	NP - 125	*Pik*c*	5	MS	4	MS
D	USEN (China)	*Pi-a+(= Pi4(t))*	7.5	S	6	S
E	Dular (Pakistan)	*Pi-ka+*	3.5	MS	2	MR
F	Kanto - 51	*Pi-k*	3.5	MS	3.5	MS
G	Shi-tia-tao (China)	*Pi-ks*	5.5	S	5	MS
H	Calaro (U. S.)	*Pi-9*	4.5	MS	4	MS
Designated race number				**ID231^{acefh}**		**ID240^{cefgh}**

Speculation on the corresponding *Avr* gene in the *Magnaporthe oryzae* population expressing race-specific interactions on international rice differentials

During the present investigation, eight differential lines carrying various known *R* genes and gene combinations exhibited specific interactions with certain lines when challenged with surviving *Magnaporthe oryzae* populations. As a result, incompatible interactions were observed (Tables- 3, 4, and 5). These incompatible interactions, typically associated with a hypersensitive response, suggest a functional interplay between avirulence (*Avr*) genes in the pathogen and corresponding *R* genes in the host.

The resistance responses induced in these differentials result from molecular cross-talk between the products of *Avr* genes and their corresponding *R* gene products. Based on the specific interactions observed in this study, the presence of corresponding *Avr* gene(s) in *M. oryzae* was inferred (Tables- 6, 7, and 8). Resistance conferred by a single major *R* gene is generally effective against races of *M. oryzae* that carry the matching *Avr* gene (Silue *et al*., 1992).

Two sets of NILs are now available, in which single major resistance genes against *M. oryzae* have been introgressed through repeated backcrossing into the genetic backgrounds of two highly susceptible rice cultivars CO39 (*indica*) and Lijiangxintuanheigu (LTH, *japonica*) (Ling *et al*., 1995; Mackill and Bonman, 1992). Within the scope of the major genes included in these sets, isolates can now be unambiguously classified into pathotypes based on their reactions to the NILs.

Following the gene-for-gene model where a single host resistance gene corresponds to one pathogen *Avr* gene in the rice blast pathosystem (Silue *et al*., 1992) the functional *Avr* gene composition of an isolate can be inferred from its pattern of incompatibility with the NILs. A nonfunctional *Avr* gene is defined by compatibility with a NIL, though it remains unclear whether the gene is absent, unexpressed, or functional in some other way. For clarity in this paper, we refer to such nonfunctional *Avr* genes as "virulences."

Despite substantial evidence supporting the gene-for-gene model in the rice–*M. oryzae* interaction, it remains an assumption that differences in compatibility between two isolates on a given NIL correspond to differences in a single *Avr* gene.

Table 6: Speculated corresponding *Avr* gene in *Magnaporthe oryzae* population expressing avirulence on international rice differential at stage I -nursery stage.

Differentials	Name of Differentials	Gene(s)	Corresponding *Avr* gene in the pathogen (*Magnaporthe oryzae*)	
			Year 2023	Year 2024
B	**Zenith** (U. S.)	*Pi-z + Pi-a + Pi-i*		*AvrPi-z + AvrPi-a + AvrPi-i*
C	**NP - 125**	*Pik*c*		*AvrPik*c*
D	**USEN** (China)	*Pi-a+(= Pi4(t))*	*AvrPi-a+*	
Race number identification			IE244^{abcgh}	II256^{adefgh}

Table 7: Speculated corresponding *Avr* gene in *Magnaporthe oryzae* population expressing avirulence on international rice differential at stage II and III during 2023.

Differentials	Name of Differentials	Gene(s)	Corresponding *Avr* gene in the pathogen (*Magnaporthe oryzae*)	
			Stage II	Stage III
C	**NP - 125**	*Pik*c*	*AvrPik*c*	NIL
E	**Dular** (Pakistan)	*Pi-ka+*	*AvrPi-ka+*	NIL

Table 8: Speculated corresponding *Avr* gene in *Magnaporthe oryzae* population expressing avirulence on international rice differential at stage II and III during 2024.

Differentials	Name of Differentials	Gene(s)	Corresponding *Avr* gene in the pathogen (*Magnaporthe oryzae*)	
			Stage II	Stage III
A	**Raminad -STR -3**	*unknown*	*Avr* gene	
B	**Zenith** (U. S.) (American cultivar Zenith)	*Pi-z + Pi-a + Pi-i*	*AvrPi-z + AvrPi-a + AvrPi-i*	*AvrPi-z + AvrPi-a + AvrPi-i*

*=Speculated Corresponding *Avr* gene in the pathogen (*Magnaporthe oryzae*)

Reaction of Monogenic Lines at nursery and Vegetative Stages to *Magnaporthe oryzae* in Jagdalpur

The monogenic lines (including an international set of differentials) were exposed to a natural blast population, and the disease assessment was done at nursery and vegetative phase at 21 and 35 DAT in 2023 and 19 and 38 DAT during 2024. Immune or highly resistant reactions of selected monogenic rice lines were observed at the nursery stage against *Magnaporthe oryzae* during 2023 and 2024. Several lines, particularly those carrying resistance genes such as *Pi1*, *Pi2*, *Pi54*, *Pi9*, *Pi-7*, and *Pi-12*, consistently exhibited immunity or high resistance across both years. (Table-9).

Table 9 : Immune / Resistant reaction of monogenic lines at nursery stage against the *Magnaporthe oryzae* population in Jagdalpur.

Line no.	Genotype	Gene	2023 Reaction	2024 Reaction
29	RP Patho-7	*Pi1*	I	I
32	RP Biopatho-1		I	I
34	RP Biopatho-3	*Pi2*	I	R
27	RP Patho-2		I	R
33	RP Biopatho-2		I	I
28	RP Patho-3	*Pi54*	I	I
37	PRS-50 (RP 6618)		R	I
39	RP 6617-59 (PRS-59)	*Pi9*	I	I
6	RIL - 29	*Pi-7*	R	R
5	RIL - 10	*Pi-12*	R	R
9	BL-245	*Pi-2 + Pi-4*	R	I
36	PRS-17	*Pi9 + Pi54*	I	R

15 out of 39 monogenic lines showed moderate resistance (MR) at the adult/vegetative growth stages against the blast pathogen in 2023 and 2024. These genotypes displayed small, slow-developing lesions, typical of "slow-blasting" resistance. Lines carrying genes like *Pi1*, *Pi2*, *Pi54*, *Pi9*, *Pi-ta*, and *Pi-kh+* showed this moderate resistance, especially under natural field conditions, highlighting the potential durability of such partial resistance mechanisms when complete resistance is absent. (Table-10)

Table 10: Moderate Resistant reaction of monogenic lines at adult / vegetative growth stage against the *Magnaporthe oryzae* population in Jagdalpur during 2024.

Line no.	Genotype	Gene	2023		2024	
			Observation Days after transplanting			
			21	35	19	38
29	RP Patho-7	*Pi1*	MR	-	R	-
32	RP Biopatho-1		MR	-	-	-
34	RP Biopatho-3	*Pi2*			MR	-
2	C101 A51		MR	-	-	-
33	RP Biopatho-2		MR	-	MR	MR
28	RP Patho-3	*Pi54*	MR	-	-	-
35	RP Biopatho-4		MR	-	-	-
39	RP 6617-59 (PRS-59)		MR	-	-	-
7	*O. minuta*	*Pi9*		-	MR	MR
22	Tetep	*Pi-kh+*		-	R	R
20	Tadukan	*Pi-ta*	MR	-	R	MR
8	BL-122	*Pi-1 + Pi-2*	MR	-	-	-
9	BL-245	*Pi-2 + Pi-4*		-	MR	MR
36	PRS-17	*Pi9 + Pi54*	MR	-	MR	MR
24	Rasi	Resistant	-	-	MR	-

2= (MR) Moderately Resistant: Small roundish to slightly elongated, necrotic gray spots, about 1-2 mm in diameter, with a distinct brown margin. Lesions are mostly found on the lower leaves; **3=(MR) Moderately Resistant**: Lesion types same as in 2 with 1-3 mm in diameter, but a significant number of lesions on the upper leaves.

Few monogenic lines that expressed a clear resistant (R) reaction at the vegetative stage in 2024. Notably, RP Patho-7 (*Pi1*), Tetep (*Pi-kh+*), and Tadukan (*Pi-ta*) were effective against the prevailing pathogen population, implying the presence of the corresponding avirulence genes (*AvrPi1*, *AvrPi-kh+*, and *AvrPi-ta*) in *M. oryzae*. These findings confirm the continued effectiveness of these specific R genes at later plant growth stages under field conditions. (Table-11).

Table 11: Resistant reaction of monogenic lines at adult / vegetative growth stage against the *Magnaporthe oryzae* population in Jagdalpur during 2024.

Line no.	Genotype	Gene	2024		*Avr gene
29	RP Patho-7	*Pi1*	R		*AvrPi1*
22	Tetep	*Pi-kh+*	R	R	*AvrPi-kh+*
20	Tadukan	*Pi-ta*	R		*AvrPi-ta*

The coevolution of plants and microbes has produced a wide spectrum of molecular interactions and host defense strategies. Plant responses to microbial attack range from full susceptibility to robust immunity, depending on the nature of the interaction and the host's genetic background (Dixon & Lamb, 1990; Keen, 1990; Long & Staskawicz, 1993). In the case of rice blast caused by *Magnaporthe oryzae*, this dynamic is further complicated by the pathogen's high genetic plasticity and the complex interplay of host resistance (R) and pathogen avirulence (*Avr*) genes.

This study evaluated the race structure of *M. oryzae* over two years in Jagdalpur using international differential cultivars and monogenic lines. The emergence of distinct races IE244^{abcgh} and II256^{adefgh} at the nursery stage, and multiple races such as IG254^{abdfh} and ID240^{cefgh} at the vegetative stages confirms that *M. oryzae* populations are temporally dynamic and capable of rapid adaptation. These findings are in line with earlier studies that reported stage-specific or seasonally variable virulence patterns due to pathogen

evolution and selection pressure (Bonman *et al*., 1987; Mackill & Bonman, 1992).

One of the major outcomes was the consistent resistance observed in monogenic lines carrying genes such as *Pi1*, *Pi2*, *Pi54*, *Pi9*, *Pi-7*, and *Pi-12*, which remained effective across years and growth stages. This confirms their continued relevance in breeding programs and aligns with prior work by Wang *et al*. (2017) and Silue *et al*. (1992), who documented their broad-spectrum effectiveness in diverse environments.

Interestingly, several genotypes displayed moderate resistance, with slower disease progression and fewer lesions. This phenotype is reminiscent of slow-blasting resistance, a form of partial, durable resistance initially described by Villareal (1980, 1981). He reported that cultivars like IRAT 13 and Gogowierie had reduced apparent infection rates and lesion sizes, suggesting a delayed or less aggressive pathogen invasion. Our observations of lines such as RP Biopatho-2 (*Pi54*), PRS-17 (*Pi9*+*Pi54*), and BL-245 (*Pi-2* + *Pi-4*) exhibiting similar patterns indicate that these traits might still be valuable under natural field pressure. Moreover, the reduction in sporulation, a key trait of slow-blasting resistance (Leonard, 1969; Villareal *et al*., 1981), further supports the idea that these moderate reactions are not merely incomplete but represent a distinct resistance strategy. These findings reinforce the value of slow-blasting as a polygenic, stable resistance model that complements major R-gene deployment and enhances the durability of blast resistance in breeding programs.

In terms of molecular basis, race-specific resistance was validated through differential responses and the inference of corresponding *Avr* genes. Incompatible reactions in lines like Zenith (*Pi-z* + *Pi-a* + *Pi-i*) and RP Patho-7 (*Pi1*) suggest the presence of their matching *Avr* genes in local pathogen populations. However, the breakdown or absence of resistance in others e.g., *Pi-a* and *Pi-k* lines indicates either virulence evolution or *Avr* gene deletion. These observations echo the work of Valent (1997) and more recently Chen *et al*. (2024), who reported frequent *Avr* gene loss or silencing in response to selection pressure from widely used R genes.

Furthermore, this study underscores the influence of environmental and physiological factors on virulence expression. The differential races observed at 21, 35, 19, and 38 days after transplanting across seasons point to host developmental stage as a modulator of resistance expression, consistent with findings by Khan *et al*. (2022) and Singh & Sharma (2023). Additionally, competition among pathogen races may shift dominance as the plant ages or environmental conditions change (Fernando *et al*., 2023). The high adaptability of *M. oryzae* is also linked to genomic plasticity mediated by transposable elements and diverse effector repertoires which has been demonstrated in recent genomic studies (Zhang *et al*., 2024; Liu *et al*., 2023).

The findings of this research, therefore, highlight the need for dynamic resistance management strategies. While strong, specific R genes remain essential tools, their effectiveness can be transient in the face of rapidly evolving pathogen populations. Integrating moderate, quantitative resistances such as slow-blasting, alongside continuous race monitoring and genomic surveillance (e.g., T2T assemblies and effector profiling), represents a more holistic and durable approach to disease management (Wang *et al*., 2024; Chen *et al*., 2024)

Conclusion

The reaction expressed by the international set of eight differential cultivars during two consecutive years (at seedling stage in the nursery on 17th August 2023 and on 31st August 2024) revealed two distinct races: IE244^{abcgh} and II256^{adefgh}. The international set of eight differential was at vegetative phase [21 days-stage II and 35 days – stage III] during 2023 and at 19 days–stage II and 38 days- stage III expressed during 2024, at which scoring of the blast disease was done. At the vegetative stage, which differs physiologically from the nursery stage, race designation varied across stages and years 2023: IG254^{abdfh} (21 days, Stage II); IB128^{ace} (35 days, Stage III); and year 2024: ID231^{acefh} (19 days, Stage II); ID240^{cefgh} (38 days, Stage III).

Based on specific interactions observed during the investigation, corresponding *Avr* genes in the pathogen were speculated, aligning with the gene-for-gene model. Resistant monogenic lines carried genes like *Pi1, Pi2, Pi54, Pi9, Pi-7, Pi-12,* and combinations of these genes, which expressed resistance or immunity to certain races. Interestingly, some lines exhibited moderate resistance (MR), characterized by small, slow-developing lesions. This phenotype, potentially linked to the "slow blasting" phenomenon, involves delayed pathogen recognition and reduced disease efficacy due to fewer, smaller lesions and decreased sporulation capacity. Such traits can limit disease spread, contributing to partial resistance and adding durability to blast resistance strategies. Understanding these race dynamics, along with the interactions between R and *Avr* genes, is crucial for designing sustainable rice breeding programs. This knowledge enables breeders to deploy resistant cultivars

strategically, mitigating the impact of evolving blast pathogen populations.

Overall, this study confirms the high virulence plasticity of *M. oryzae* and the effectiveness of certain R genes (*Pi1*, *Pi2*, *Pi54*, *Pi9*) under dynamic field conditions. The detection of moderate resistance linked to slow-blasting traits adds a new dimension to breeding strategies, suggesting that polygenic, durable resistance should be prioritized over reliance on major, race-specific R genes alone. Ongoing virulence monitoring, along with genomics-based *Avr* gene tracking, is critical for sustainable rice blast management.

Acknowledgment

I gratefully acknowledge the support provided by the Department of Science and Technology (DST), Government of India, through the INSPIRE Fellowship. This fellowship has been instrumental in facilitating my research and academic progress. I would also like to thank Dr. R.S. Netam, Dean, SGCARS, Jagdalpur, for his generous support and for ensuring access to the necessary research facilities in Jagdalpur.

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