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PHENOTYPIC SCREENING AND MOLECULAR CHARACTERISATION OF ADVANCED BREEDING LINES FOR INTROGRESSION OF BLAST RESISTANT GENES IN RICE (*ORYZA SATIVA* L.)

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ABSTRACT

Biotic stresses are major threat to rice production. Among the biotic stresses, blast is one of the major diseases affecting rice grain production. Present investigation was conducted to evaluate genotypic and phenotypic effect of 50 breeding lines from a cross (MTU1010 x Akshayadhan NIL) in glass house at Regional Agricultural Research Station, Jagtial. 50 advanced breeding lines were screened through foreground selection for confirmation of presence of target trait specific genes *viz*., for blast (*Pi54*) was carried out with co-dominant marker *Pi54*-MAS for checking the presence of *Pi54*. Out of 50 advanced breeding lines, 35 lines possess resistant allele of *Pi54* gene. Phenotypic screening of 50 breeding lines was done in uniform blast nursery; among them 35 breeding lines were shown resistance against blast disease. The results obtained in the present study indicate the success of combining marker assisted breeding with phenotypic selection. Cultivation of biotic stress resistance breeding lines developed in this study would be great advantage in blast endemic areas and can be expand the area under MTU1010 NIL.

Keywords **:** Rice; Blast; *Pi54*; MTU1010 NIL and Akshayadhan NIL

Introduction

Rice (*Oryza sativa* L.) is one of the most important food crops that helps in the survival of more than 50% of the world population. It played a significant role in food security of Asia, where more than 90 per cent of the rice is produced and consumed. The rice productivity is influenced by numerous biotic and abiotic factors. Among biotic stresses, fungal diseases are of major concern causing significant yield losses in the rice crop throughout the world. Amongst diseases, rice blast caused by *Magnaporthe oryzae* is one of the most destructive diseases in rice. The blast pathogen is a heterothallic fungus and diverse phenotypic virulence develops through evolution of new races. Wide geographic distribution, continuous evolution of new races, and high yield losses make this fungal disease a severe threat to rice production. It is estimated that every year rice blast is responsible to cause 10-30% yield losses in rice that resulted in severe epidemics in major rice growing regions of the world. Although, the fungicide application provided a short-term solution but it is practically uneconomical for resource poor farmers. Utilization of broad-spectrum blast resistant varieties is the most successful, economical and environmental responsive approach to manage blast disease. Disease management approach required the better understanding of the phenotypic and genetic diversity of the *M. oryzae* populations that

would help in devising the strategies to manage this disease.

As on date, 118 blast major rice resistance genes (R-genes) have been identified Yadav *et al*. (2019). Among the major blast resistance genes, *Pi-k*^{h}, which has been recently renamed as *Pi54* Sharma *et. al*. (2010), exhibited resistance to predominant races of the pathogen in India Sharma *et al*. (2002). *Pi54* gene was originally identified in Tetep, a Vietnam's *indica* variety and mapped on chromosome 11L with two tightly linked simple sequence repeat (SSR) markers TRS26 and TRS33 has been cloned Sharma *et al*. (2005). Genotyping using these linked markers requires analysis through Poly Acrylamide Gel Electrophoresis (PAGE), which is cumbersome and time-consuming. Hence, marker-assisted introgression of *Pi54* into susceptible rice varieties is being achieved through another linked SSR marker, RM206, which can be resolved through agarose gel electrophoresis Srinivasarao *et al*. (2009) and Hari *et al*. (2013). However, the marker is not very close to the gene and this might result in some recombinants in marker assisted selection. Ramkumar *et al*. (2011) developed PCR based functional marker *Pi54* MAS was observed to perfectly co- segregate with no recombinants. The rice cultivar Tetep has been found to be resistant to most of the pathogenic races occurring in India (Padmanabhan, 1979). NLR 34449 (Nellore mahsuri) a blast resistant variety were developed using Tetep as one of its parents.

Many popular varieties and hybrids in almost all crops were developed in Telangana and Andhra Pradesh, India among them the popular rice varieties *viz*., Cottondora Sannalu (MTU 1010), Samba Mahsuri (BPT 5204), Swarna (MTU 7029) have occupied more than 25% rice area in India. MTU 1010 (Cottondora Sannalu), a short duration mega rice variety with long slender grain released by Andhra Pradesh Rice Research Institute (APRRI), Maruteru was occupied maximum area in Andhra Pradesh and Telangana possessing BPH resistance and gives 6-6.5 tonnes/ha under good agronomic conditions particularly during *Rabi* season (APPRI, Maruteru). However, it is susceptible to blast diseases. Hence there is an urgent need to improve this mega variety by incorporating resistance genes of blast.

The present investigation has been taken with an aim to introgress blast resistance gene (*Pi54*) from Akshayadhan into MTU 1010.

Material and Methods

Plant material

ICAR-Indian Institute of Rice Research, Hyderabad has developed a high yielding variety, Akshayadhan, by crossing an *Indica* breeding line and a Tropical *Japonica* derivative. Even though the variety recorded very high yield advantage, it is susceptible to blast disease. Therefore, ICAR-IIRR has developed NILs of Akshayadhan (RP 6132) with blast resistance (*Pi54*) through marker-assisted backcross breeding Bhaskar *et al*. (2015). NIL of Akshayadhan (RP 6132) having *Pi54* genes was used as donor parent/male parent. MTU 1010 (IET 15644) is a short duration semi dwarf variety (120-125 days) high yielding, developed from cross between Krishnaveni and IR 64 it was used as a recurrent parent. In addition to these, Taichung Native 1 (TN1) was used as a susceptible check and NLR 34449 was used as resistant check for blast screening of advanced breeding lines.

SSR markers

In rice, for genetic analysis studies simple sequence repeats (SSRs) are the DNA markers are of best choice because of their abundance, high polymorphism and simple assays using agarose gel electrophoresis. Hence gene linked SSR markers were used in the present study for foreground selection.

Target gene	Chr. no	Primer Name	Primer Sequence		Annealing temp (°C)	Amplicon size	Reference
Pi54	11	*Pi54* MAS	F	CAATCTCCAAAGTTTTCAGG	55	254 (Donor allele) 350 (Recipient allele)	Ram kumar *et. al*. (2011)
			R	GCTTCAATCACTGCTAGACC			

Screening for blast resistance: The selected fifty advanced breeding lines of F_7 generation along with parents MTU1010, Akshaydhan NIL, NLR 34449 (resistant check) and TN 1 (susceptible check) were screened for blast resistance under *in-vivo* conditions *i.e*., uniform blast nursery bed at Regional Agriculture Research Station (RARS), Jagtial during *Rabi* 2020-21 (Figure-1). Pathogen strains were cultured and stored

as described by Prasad *et al*. (2011). Disease development was ensured by planting seedlings with spreader rows of susceptible variety TN1 at RARS, Jagtial and each test entry was sown in a single row of 50 cm long and 10 cm apart. After every 10 test entries susceptible check was sown. The entries are inoculated with blast suspension spore culture. Inoculated seedlings were monitored fifteen days after inoculation the test entries (Advanced breeding lines) and disease scoring was done as per the standard evaluation system (SES) 0-9 scale IRRI, 2013, Table-1.

Table 1 : Standard Evaluation System, IRRI -2013 scale for Blast disease scoring in Rice

Scale	Disease severity	Host response
0	Lesions are not present	Resistance (R)
1	Small brown specks of pin point size or large brown specks without sporulating centre	Resistance (R)
2	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	Resistance (R)
3	Lesions type is same as in scale 02, but a significant number of lesions on upper leaf area	Resistance (R)
4	Typical susceptible blast lesions, 3mm or longer infecting less than 4% of leaf area	Moderately Resistance (MR)
5	Typical susceptible blast lesions infecting 4-10% of leaf area	Moderately Resistance (MR)
6	Typical susceptible blast lesions infecting 11-25% of the leaf area	Moderately Susceptible (MS)
7	Typical susceptible blast lesions infecting 26-50% of the leaf area	Susceptible (S)
8	Typical susceptible blast lesions infecting 51-75% of the leaf area and many leaves are dead	Susceptible (S)
9	More than 75% of the leaf area affected	Susceptible (S)

Marker-assisted selection for blast resistance

For targeted introgression of *Pi54* into MTU1010, a marker-assisted pedigree breeding programme was adopted. DNA was isolated from the parents and breeding lines by following the protocol of Zheng *et al*. (1995). The PCR-based SSR marker *Pi54* MAS Ram Kumar *et. al*. (2011) was used. PCR analysis was done using gene linked markers for *Pi54* gene. PCR analysis was done to identify the genes by using forward and reverse microsatellite primers, reactions were performed in a final volume of 10µl. 3µl of diluted total genomic DNA, 1.0µl of 10X PCR buffer (10mM Tris-HCl pH 9.0, 50mM KCl, 1.5 mM $MgCl_2$ and 0.01% gelatin), 0.5 µl of dNTPs solution (2.5mM), 0.5 µl of each of forward and reverse primers (5 PM), 0.5 µl of 1.0 U Taq DNA polymerase (Banglore genei Private Limited, Banglore) and 4 µl of sterilized double distilled water. Amplification was carried out using programmable Thermal cycler (G- Strom and Eppendorf).

Results and Discussions

Genotypic Characterization of Advanced Breeding Lines for Introgression of Blast Gene *Pi54*

Screening with molecular markers is accurate and easy compared to the phenotypic screening, employment of molecular markers in breeding programmes greatly increases the efficiency of selection especially through foreground selection in combination with or to replace phenotypic screening for the target gene Collard and Mackill. (2008). DNA markers closely linked to a blast resistance gene that confers resistance to a particular race of the pathogen can be effectively employed for marker assisted selection (MAS), which is much faster than traditional phenotyping screening. A total number of 50 advanced breeding lines were genotyped for the target disease *i.e*., blast resistance (*Pi54*) in the present study, during *Rabi* 2020-21. The pooled DNA from each advanced breeding lines screened with closely linked functional marker *Pi54*-MAS Ramkumar *et al*. (2011), specific for *Pi54* through foreground selection. Among 50 advanced breeding lines 35 lines viz., VSR-1, VSR-2, VSR-3, VSR-4, VSR-5, VSR-6, VSR-7, VSR-8, VSR-9, VSR-11, VSR-19, VSR-20, VSR-21, VSR-22, VSR-23, VSR-24, VSR-25, VSR-26, VSR-27, VSR-28, VSR-29, VSR-32, VSR-35, VSR-36, VSR-37, VSR-38, VSR-39, VSR-40, VSR-41, VSR-42, VSR-43, VSR-44, VSR-45, VSR-46 and VSR-47 were identified homozygous positive for *Pi54* target trait and remaining 15 breeding lines *viz*., VSR-10, VSR-12, VSR-13, VSR-14, VSR-15, VSR-16, VSR-17, VSR-18, VSR-30, VSR-31, VSR-33, VSR-34, VSR-48, VSR-49 and VSR-50 were found homozygous negative for *Pi54*. A gel picture showing amplification

of *Pi54*-MAS marker in advanced breeding lines is given in Figure-2. Similar results were observed with Abhilash Kumar *et al*. (2016), Ellur *et al*. (2016), Laxmi Prasanna *et al*. (2018) and Rekha *et al*. (2018) and Laxmi Prasanna *et al*. (2022) and these lines may perform good in the field conditions.

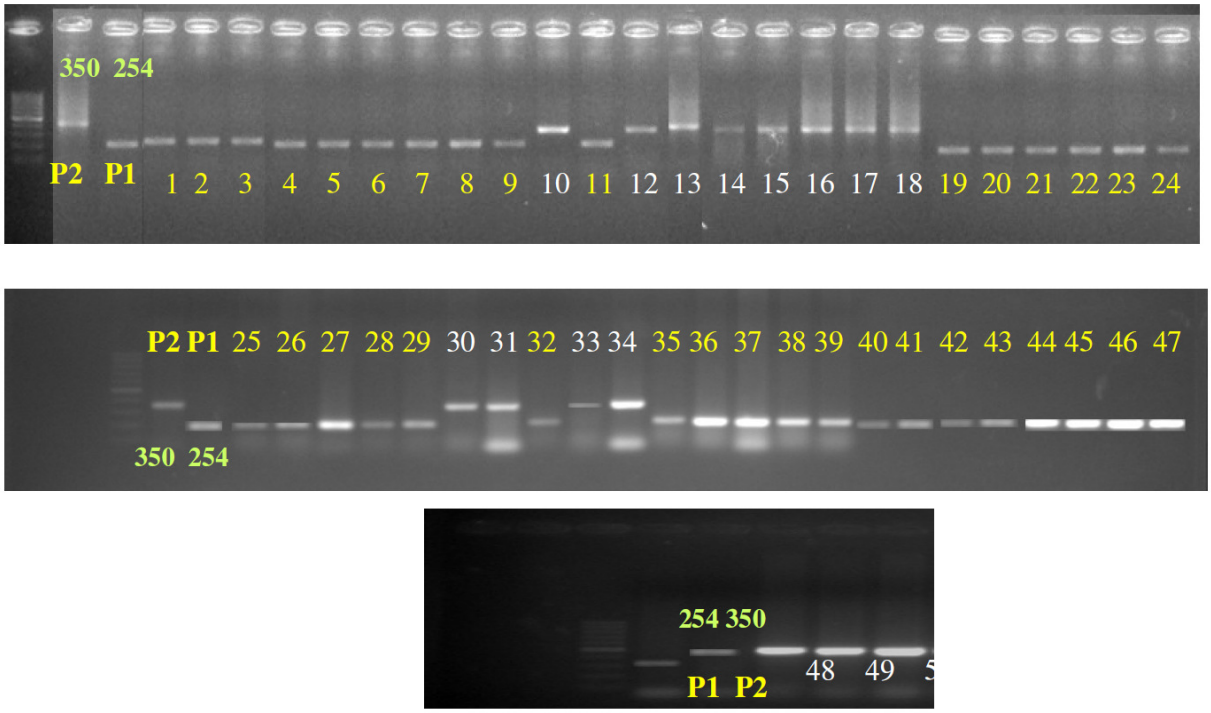

Fig. 2 : Foreground selection for target gene using a closely linked marker *Pi54*MAS for *Pi54* gene Ladder- 100 bp, P1- Akshayadhan, P2- MTU1010.

Phenotypic Evaluation of Advanced Breeding Lines for Blast

A total of 50 advanced breeding lines along with the parents and checks were screened for blast resistance under *in-vivo* conditions in Uniform blast nursery (UBN) at Regional Agriculture Research Station (RARS), Jagtial, Telangana during *Rabi* 2020-2021. A local isolate of *Magnoporthe oryzae*, named SPI-40 was used for screening the selected advanced breeding lines NLR 34449 was used as resistant check and TN1 was used as susceptible check. The plants were scored and evaluated on a 0-9 scale as per 5th edition of SES (IRRI, 2013). Among the 50 advanced breeding lines screened for blast resistance, 35 breeding lines *viz*., VSR-1, VSR-2, VSR-3, VSR-4, VSR-5, VSR-6, VSR-7, VSR-8, VSR-9, VSR-11, VSR-19, VSR- 20, VSR-21, VSR-22, VSR-23, VSR-24, VSR-25, VSR-26, VSR-27, VSR-28, VSR-29, VSR- 32, VSR- 35, VSR-36, VSR-37, VSR- 38, VSR-39, VSR-40, VSR-41, VSR-42, VSR-43, VSR-44, VSR-45, VSR-46 and VSR-47 were found resistant with a disease score of three (3), 14 breeding lines *viz*., VSR-10, VSR-12, VSR-13, VSR-14, VSR-15, VSR-16, VSR-17, VSR-18, VSR-30, VSR-31, VSR-33, VSR-34, VSR-48 and VSR-50 observed to be moderately resistant with a disease score of 5. The remaining breeding line VSR-49, were found susceptible with a disease score of 7. The susceptible check, TN1 and the parent MTU1010 were highly susceptible to blast with a score of 9. The resistant check, NLR34449 was found highly resistant to blast disease with score of 1. The parent Akshayadhan showed resistant reaction with a score of 3 in the Table-2. All the advanced breeding lines showing resistance to both bacterial blight and blast were advanced to observational varietal trial.

Pyramiding of two or more genes found effective in the cultivars Sundaram *et al*. (2008), Usatov *et al*. (2016) and Rekha *et. al*. (2018) than the single gene conferred resistance, many studies have shown that the major blast resistance gene conferred durable resistance in the studies Hari *et al*. (2013), Abhilash *et al*. (2016), Laxmi Prasanna *et al*. (2018), Swathi *et al*. (2019).

Table 2 : Screening details Advanced breeding lines for blast resistance and scoring details as per IRRI-SES scale (IRRI, 2013)

Parents and Checks	Reaction against Blast SPI-40	
	Score	R/MR/S
MTU1010 NIL	9	S
Akshayadhan NIL	3	R
NLR 34449 (Resistant Check)	1	R
TN1 (Susceptible check)	9	S
Improved breeding lines	Score	R/MR/S
VSR 1	3	R
VSR 2	3	R
VSR 3	3	R
VSR 4	3	R
VSR 5	3	R
VSR 6	3	R
VSR 7	3	R
VSR 8	3	R
VSR 9	3	R
VSR 10	5	MR
VSR 11	3	R
VSR 12	5	MR
VSR 13	5	MR
VSR 14	5	MR
VSR 15	3	R
VSR 16	5	MR
VSR 17	5	MR
VSR 18	5	MR
VSR 19	3	R
VSR 20	3	R
VSR 21	3	R
VSR 22	3	R
VSR 23	3	R
VSR 24	3	R
VSR 25	3	R
VSR 26	3	R
VSR 27	3	R
VSR 28	3	R
VSR 29	3	R
VSR 30	5	MR
VSR 31	5	MR
VSR 32	3	R
VSR 33	5	MR
VSR 34	5	MR
VSR 35	3	R
VSR 36	3	R
VSR 37	3	R
VSR 38	3	R
VSR 39	3	R
VSR 40	3	R
VSR 41	3	R
VSR 42	3	R
VSR 43	3	R
VSR 44	3	R
VSR 45	3	R
VSR 46	3	R
VSR 47	3	R
VSR 48	5	MR
VSR 49	7	S
VSR 50	5	MR

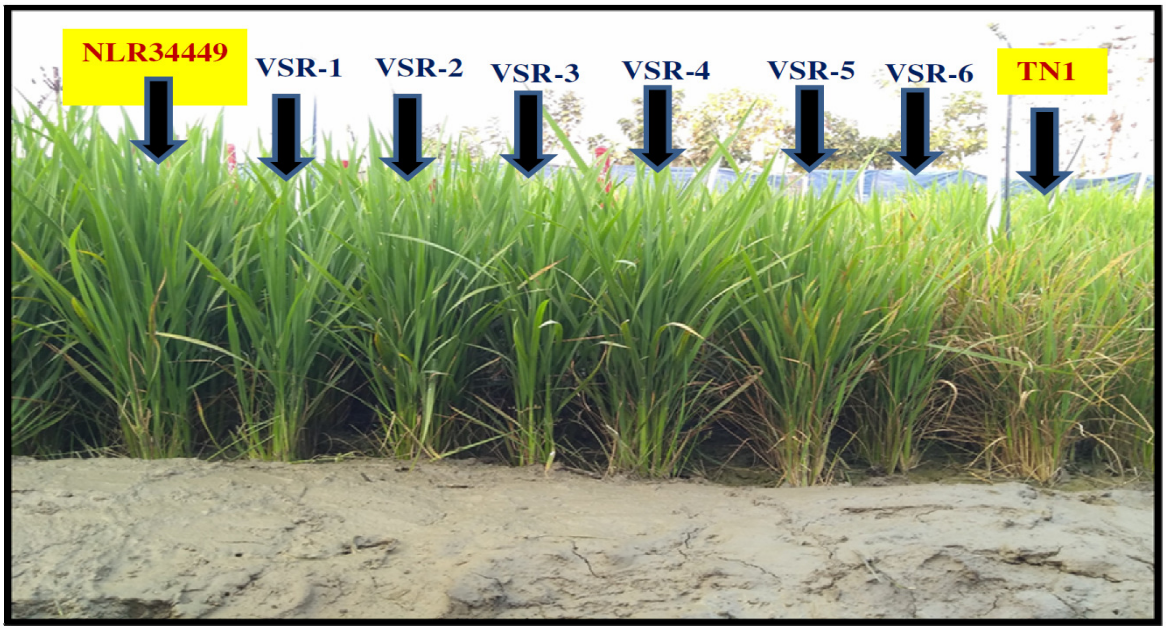

Fig. 1 : Phenotypic screening for blast resistance using *Magnaporthe grisea* culture. NLR34449: Resistant check; TN1: Susceptible Check; VSR-1 to VSR-6 Advanced breeding lines possessing *Pi54* gene.

Conclusion

Superior breeding lines developed in the present study (Resistant and moderately resistant lines) will be advanced and could be evaluated through multilocation trails in the state of Telangana and also across the country through All Indian Coordinated Research Improvement Project (AICRP). After the evaluation of these breeding lines in the field condition, then can be evaluated through the above-mentioned trials and the best one can be released as a variety to serve the purpose of farmer's need in the state. Also, these breeding lines possessing blast resistance along with good yield levels can be serve as good donors for targeted transfer of the major gene to other elite rice varieties cultivated in Telangana state.

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References

Abhilash, K.V., Balachiranjeevi, C.H., Bhaskar, N.S., Rambabu, R., Rekha, G., Madhav, K.R., Vijay, S., Pranathi, K., Harika, G., Mahadevaswamy, H.K., Anila, M., Hajira, S.K., Yugander, A., Hariprasad, A.S., Madhav, M., Laha, G.S., Balachandran, S.M., Sundaram, R.M and Prasad, M.S. (2016). Marker-assisted introgression of bacterial blight and blast resistance genes into RPHR 1005, restorer line of the popular rice hybrid, DRRH-3. *Journal of Plant Biochemistry and Biotechnology* **25**(4): 400–409.

Bhaskar, N.S., Balachiranjeevi, C.H., Abhilash, V., Harika, G., Laha, G.S., Prasad, M.S and Sundharam, R.M. (2015). Introgression of bacterial blight and blast resistance into the elite rice variety, Akshayadhan through marker assisted backcross breeding. *International Journal of Current Research* **7**(8): 18943-18946.

Collard, B.C and Mackill, D.J. (2008). Marker assisted selection an approach for precision plant breeding in the twenty-first century. *Philosophical Transactions of the Royal Society B Biological Sciences* **363**(1491): 557-572.

Ellur, R.K., Khanna, A., Yadav, A., Pathania, S., Rajashekara, H., Singh, V.K., Gopalakrishnan, S., Bhowmick, P.K., Nagarajan, M., Vinod, K.K., Prakash, G., Mondal, K.K., Singh, N.K., Prabhu, K.V and Singh, A.K. (2016). Improvement of Basmati rice varieties for resistance to blast and bacterial blight diseases using marker assisted backcross breeding. *Plant Science* **242**: 330-334.

Hari, Y., Srinivas Rao, K., Viraktamath, B.C., Hariprasad, A.S., Laha, G.S., Ilyas, A.M., Natarajkumar, P., Sujatha, K., Prasad, M.S., Rani, N.S., Balachandra, S.M., Kemparaju, Mohan, K.M., Sama, V.S.A.K., Shaik, H., Balachiranjeevi, Ch., Pranathi, K., Reddy, G.A., Madhav, M.S and Sundaram, R.M. (2013). Marker assisted introgression of bacterial blight and blast resistance into IR 58025b, an elite maintainer line of rice. *Journal of Plant Breeding* **132** (6): 586-594.

Laxmi Prasanna, B., Dangi, K.S., Sundaram, R.M., Damodar Raju, C.H., Jagadeeshwar, R., Rekha, G and Sinha, P. (2018). Screening of breeding lines of MTU1010 for their resistance against bacterial blight and blast. *International Journal of Current Microbiology and Applied Sciences* **7**(7): 4077-4084.

Laxmi Prasanna, B., Dangi, K.S., Damodar Raju, C.H., Jagadeeshwar, R., Rekha, G., Pragya Sinha., Aleena, D., Kousik, M.B.V.N., Mahadeva Swamy, H.K., Anila, M., Dilip kumar, T., Punniakotti, E., hajira, S.K., Bhadana, V.P and Sundaram, R.M. (2022). Marker assisted pedigree breeding based improvement of Indian mega variety of rice MTU1010 for resistance against bacterial blight and blast and tolerance to low soil phosphorous. *PLoS ONE* **17**(1): e0260535.

Padmanabhan, S.Y. (1979). Blast resistance in India. *Rice Blast Workshop*. International Rice Research Institute, Philippines,43-61.

Prasad, M.S., Sheshu, M.S., Laha, G.S., Lakshmi, D., Krishnaveni, D., Mangrauthia, S.K., Balachandran, S.M., Sundaram, R.M., Arunakanthi, B., Madhan Mohan, K., Ratna Madhavi, P., Kumar,V and Viraktamath, B.C. (2011). Rice blast disease and its management. *Technical Bulletin No. 57*. Directorate of Rice Research (ICAR), Hyderabad,52.

Ramkumar, G., Rao, S.K., Madhan, M.K., Sudarshan, I., Sivaranjani, A.K.P., Krishna, G.K., Neeraja, C.N., Balachandran, S.M., Sundaram, R.M., Prasad, M.S., Shobha Rani, N., Ram Prasad, A.M., Virakma, B.C. and Madhav, M.S. (2011). Development and validation of functional marker targeting an indel in the major rice blast disease resistance gene *Pi54 (Pik^h)*. *Molecular Breeding* **27**: 129-135.

Rekha, G., Kumar, A.V., Virakthamath, B.C., Pranathi, K., Koushik, M.B.V.N., Prasanna, L.B., Backiyalakshmi, C., Sinha, P., Ravindra, R.K., Bhaskar, S., Hajira, S.K., Balachiranjeevi, C.H., Swapnil, K., Rambabu, R., Harika, G., Punniakoti, E., Anila, M., Dilip kumar, T., Yugander, A., Chaitra, K., Praveen, M., Prasad, M.S., Laha, G.S., Neeraja, C.N., Giri, A., Subbarao, L.V., Babu, R.V and Sundharam, R.M. (2018). Improvement of blast resistance of the popular high yielding medium slender-grain type, bacterial blight resistant rice variety, improved Samba Mahsuri by marker assisted breeding. *Journal of Plant Biochemistry and Biotechnology* **27** (4): 463-472.

Sharma, T.R., Chauhan, R.S., Singh, B.M., Paul, R., Sagar, V and Rathore, R. (2002). RAPD and pathotype analysis of *Magnaporthe grisea* population from North-western Himalayan region of India. *Journal of Phytopathology* **150**: 649-656.

Sharma, T.R., Madhav, M.S., Singh, B.K., Shanker, P., Jana, T.K., Dalal, V., Pandit, A., Singh, A., Gaikwad, K., Upreti, H.C and Singh, N.K. (2005). High resolution mapping, cloning and molecular characterization of *Pikh* gene of rice., which confers resistance to *M. grisea*. *Molecular Genetics and Genomics* **274**: 569-578.

Sharma, T.R., Rai, A.K., Gupta, S.K and Singh, N.K. (2010). Broad spectrum blast resistance gene Pikh cloned from the rice line tetep designated as *Pi54*. *Journal of Plant Biochem Biotechnol* **19**:1.

Srinivasarao, K., Hari, Y., Laha, G.S., Virakthmath, B.C., Mishra, B., Hariprasad, A.S., Natarajkumar, P., Mohan, K.M., Prasad, M.S and Sundaram, R.M. (2009). Marker assisted introgression of bacterial blight and rice blast resistance genes into hybrid rice parental lines. *Proceedings of 6th International Rice Genetics Symposium*, IRRI., Philippines,53-56.

Sundaram, R.M., Vishnupriya, M.R., Sunil, K.B., Gouri, S.L., Ashok Reddy, G., Shobha Rani, N., Sharma, P.N and Ramesh V.S. (2008). Marker assisted introgression of bacterial blight resistance in Samba Mahsuri, an elite *indica* rice variety. *Euphytica* **160** (3): 411-422.

Swathi, G., Durga Rani, Ch.V., Jamaloddin, Md., Madhav, S. M., Vanisree, S., Anuradha, Ch and Jagdeeswar, R. (2019). Marker assisted introgression of the major bacterial blight resistance genes, *Xa21* and *xa13* and blast resistance gene, *Pi54,* into the popular rice variety, JGL1798. *Molecular Breeding* **39**: 58.

Usatav, A.V., Koatylev, P.I., Azarin, K.V., Markin, N.V., Makarenko, M.S., Khachumov, V.A., Bibov, M and Yu. (2016). Introgression of the rice blast resistance genes *Pi1*, *Pi2* and *Pi33* into Russian rice varieties by marker-assisted selection. *Indian Journal of Genetics and Plant Breeding* **76** (1): 18-23.

Yadav, M.K., Aravindan, S and Rath, P.C. (2019). Blast resistance in Indian rice landraces: Genetic dissection by gene specific markers. *PLoS One* **14** (3): 0213566.

Zheng, K., Subudhi, P.K., Domingo, J., Magpantay, G and Huang N. (1995). Rapid DNA isolation for marker assisted selection in rice breeding. *Rice Genetic Newsletter* **12**: 255–258.