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Screening of F_5 Families of Rice Against Resistance to *Rhizoctonia Solani* and *Xanthomonas Oryzae Pv. Oryzae*

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Abstract: *Rice is the major food crop for millions of population in the world but its production is limited by several biotic stresses of which sheath blight and bacterial leaf blight are two major devastating diseases that seriously limits rice yield and hence development and selection of resistant varieties is very much needed. In this context, in the present study thirty five F_5 families of rice obtained from the* two *crosses MTU 7029/ NLR 34449 and MTU 7029/ PAU 3116-25-5-1 were screened against sheath blight by adopting typha leaf bit method of artificial inoculation followed by field screening using 0-9 scale of SES, 2014. Same families were also screened against bacterial leaf blight where leaf clipping method is used for artificial multiplication of inoculums. Results revealed that 18 families had reported moderate resistance to sheath blight whereas only seven families showed moderate resistance to bacterial leaf blight, hence can be selected. Five families recorded moderate resistance to both the diseases and hence more preferred.*

Keywords: *F5 familes, sheath blight, bacterial leaf blight, typha leaf bit, leaf clipping.*

1. Introduction

Rice is a major cereal crop that contributes significantly to global food security. Rice provides 21% of global human per capita energy and 15% of per capita protein. Paddy cultivation suffers from several biotic and abiotic stresses that seriously affect its production among which Sheath blight, caused by *Rhizoctonia solani* (teleomorph: *Thanatephorus cucumeris*) and Bacterial leaf blight, caused by *Xanthomonas oryzae pv. Oryzae* were the two major devastating dieases in many countries affecting more than 50% of global rice production (Khush and Ogawa, 1989 and Marchetti and Bollich, 1991).

Sheath blight is a fungal disease caused by Rhizoctonia solani. Symptoms are usually observed from tillering to milk stage in a rice crop. Rice sheath blight is found in all rice production areas, and is decreasing rice production especially in intensified production systems. It spreads through sclerotia present in the soil which develops primary mycelium with the onset of favourable conditions that forms initial lesions on sheath which later develops into runner hyphae that grow on the surface of rice plant tissues, and develop infection structures that generate new lesions. Disease intensification and spread are also favoured by long duration of tissue wetness, crop canopy and canopy microclimate. The fungus affects the crop from tillering to heading stage. Initial symptoms are noticed on leaf sheaths near water level. On the leaf sheath oval or elliptical or irregular greenish grey spots are formed. As the spots enlarge, the centre becomes greyish white with an irregular blackish brown or purple brown border. Lesions on the upper parts of plants extend rapidly coalesing with each other to cover entire tillers from the water line to the flag leaf. The presence of several large lesions on a leaf sheath usually causes death of the whole leaf, and in severe cases all the leaves of a plant may be blighted in this way. The infection extends to the inner sheaths resulting in death of the entire plant. Older plants are highly susceptible. Five to six week old leaf sheaths are highly susceptible. Plants heavily infected in the early heading and grain filling growth stages produce poorly filled grain, especially in the lower part of the panicle. Many rice cultivars have been identified as moderately resistant to sheath blight, however no resistant cultivar has been found so far (Prasad and Eizenga, 2008).

Bacterial blight is a monsoon disease and its incidence and severity is very much influenced by rainfall, number of rainy days, and susceptibility of the cultivar and nitrogen fertilizer application. Severe epidemics recorded in 1979 and 1980 in northwestern India, reducing the grain yields drastically.

The bacterium induces either wilting of plants or leaf blight. Wilting syndrome known as 'Kresek' occurs sporadically in the fields causing serious damage. It commonly occurs within 3-4 weeks after transplantation of the crop. Kresek results either in the death of whole plants or wilting of only a few leaves. Leaf blight phase is the most predominant form of the disease occurring between tillering and heading stage of the corp. Third type of less conspicuous symptoms caused by the bacterium is yellowing of leaves. Such leaves show blighted appearance.

The earliest symptom of the blight phase is the appearance of dull greenish water-soaked or yellowish spots 5-10 mm in length on the leaf towards the tip or margins, leading to tip and marginal dying. The infection soon extends along one or both margins, sometimes to the leaf sheath. As the disease progress, several lesions coalesce to form straw-brown large lesions or blighted portions. The inner margin of blighted patch in contact with the adjoining green portion of the leaf is ragged or wavy. Occasionally, the lesion may extend from the tip downward along the midrib itself, the leaf margins remaining green. Small droplets of bacterial ooze, pale amber in colour, are found on the affected portions. Lesions may also be seen on leaf sheaths in susceptible varieties. Milky or opaque dew drops containing bacterial masses are formed on young lesions in the early morning. They dry up on the surface leaving a white encrustation. The affected grains have discoloured spots surrounded by water soaked areas. If the cut end of leaf is dipped in water, bacterial ooze makes the water turbid.

The most effective approach to control these two diseases is using resistant varieties. Development of disease resistant rice is one of the most important achievements rice breeders attempt to accomplish. The genetic diversity of rice may incorporate genes that directly contribute to physiological host plant resistance to sheath blight (Srinivasachary et al. 2011), genes that determine the architecture of plants, and thus contribute to the structure of crop canopies, as well as genes from these different groups that collectively confer resistance through interactions which can be identified by field screening by standardized methods. Hence, the objective of the present study therefore was to screen thirty five F5 families of rice against sheath blight and bacterial leaf blight which will enable us to identify rice varieties resistant to these diseases.

2. Material and Methods

In the present study, during *kharif* 2015, all the thirty five F_5 families obtained from two crosses MTU 7029/ PAU 3116-25-5-1 and MTU 7029/ NLR 34449 along with their susceptible check (MTU 7029) were sown in two rows each with a spacing of 20 x 15 cm at Andhra Pradesh Rice Research Institute and Regional Agricultural Research Station, Maruteru, West Godavari District, Andhra Pradesh and were screened against sheath blight by adopting typha leaf bit method of artificial inoculation done at 69 DAS followed by field screening at maximum tillering stage and panicle initiation stage when 95% of check variety was affected using 0-9 scale of Standard Evaluation System give by IRRI, 2014 (Table 1). These families were also screened separately for bacterial leaf blight by following leaf clipping method for artificial multiplication of bacteria at 80 DAS followed by field screening at maximum tillering stage and later at panicle initiation stage when 95% of check variety TN-1 was affected using 0-9 scale of Standard Evaluation System (SES) of IRRI, 2014 (Table 1). Fertilizer management and plant protection measures for other pests and diseases were followed as per recommendations.

Screening for disease resistance based on natural infection may not always be conclusive due to environmental variation and the absence of adequate inoculum that initiates the disease. Artificial inoculation minimizes such problems. Hence, for proper infestation and to get good reaction of test seedlings, artificial inoculation by typha leaf bit method for sheath blight and leaf clipping method for bacterial leaf blight were practiced in the present study.

2.1 Typha Leaf Bit Method

This method was first used by Bhaktavastalam *et al.* (1978) for mass multiplication of Sheath blight causing fungus. In this method, uniform sized typha bits were cut and sterilized in autoclave and inoculated with *Rhizoctonia solani*. The material is kept under wet condition for multiplication of the fungus. After complete coverage of the typha bits with fungal mat, the bits were used for artificial inoculation. Two bits per hill were used for artificial inoculation. The bits were inserted in between the tillers at the base of the plant and tied with thread so as to come in contact with the neighbouring tillers. Inoculated hills were observed for the appearance of the symptoms twice, initially at maximum tillering and later at panicle initiation stages and scores were recorded as per 0-9 scale of SES, IRRI, 2014. Highest score among the two was considered as final one.

2.2 Leaf Clipping Method

Kauffman et al. (1973) reported the leaf clipping method of artificial inoculation for bacterial leaf blight disease. In this method, sterilized surgical scissors dipped in bacterial suspension were used for inoculation. Leaves of all the three plants in a pot were grasped in one hand and the top 1-3 inches of three leaves were clipped off simultaneously. The inoculum should be used within two hours after preparation as Xanthomonas oryzae pv. oryzae quickly losses its viability. A control of each variety was also maintained, by using scissors dipped in sterile water for clipping off the leaves. This method is very efficient for inoculating large amount of breeding materials in the field and is currently being used at IRRI, Phillippines. One should note that in both seedling and field tests, folded young leaves and old leaves or leaves with symptoms of nutrient deficiency or other diseases should be avoided for inoculation.

3. Results and Discussion

Screening was conducted at Andhra Pradesh Rice Research Institute, Maruteru during kharif, 2015 based on SES, IRRI, 2014. All forty three F5 families were grouped into five classes based on their susceptibility to that disease viz., immune or highly resistant with score 1, resistant with score 3, moderately reistant with score 5, susceptible with score 7 and highly susceptible with score 9.

Immunity refers to the inability of the pathogen to cause disease symptoms on host plant. No yield loss will be observed in this case where as Resistance refers to the ability of a plant to overcome completely or in some degree the effect of a pathogen or damaging factor. Yield loss is very low or neglible when seen in economic terms. Moderately reistant plants can tolerate disease to some extent giving moderate to high yield when disease intensity is low where as suscetibility refers to inability of a plant to resist the effect of a pathogen or other damaging factor. yield loss will be high in this case. Highly susceptible plants cannot withstand lower intensity of disease and complete yield loss will be observed under such circumstances. Practically it is very difficult to develop immune varieties. Hence plant breeders mostly concentrate on developing resistant and moderately resistant varieties.

In the present study, among the thirty five F5 families screened against sheath blight and bacterial leaf blight, no family was found to be immune or resistant. In the F5 population obtained from the cross MTU 7029/ NLR 34449, among the twelve F5 families, five recorded moderate resistance (score 5) while seven were susceptible to sheath blight (score 7) where as two families recorded moderate resistance (score 5) while nine were susceptible (score 7) to bacterial leaf blight. (Table I).

TABLE I: Standard Evaluation System, IRRI (2014) scale for sheath blight

Scale for Sheath blight		
Scale	**Rating**	**Disease symptoms**
0	Highly Resistant	No Infection (Immune reaction)
1	Resistant	Lesions limited to lower 20% of the plant height
3	Moderately Resistant	20-30
5	Susceptible	31-45
7	Highly Susceptible	46-65
9	Highly Resistant	>65
Scale for Bacterial leaf blight		
Scale	**Rating**	**% leaf area diseased**
1	Highly Resistant	1-5
13	Resistant	6-12
5	Moderately Resistant	13-25
7	Susceptible	26-50
9	Highly Susceptible	51-100

Out of twenty three F5 families obtained from the cross MTU 7029/ PAU 3116-25-5-1, thirteen families recorded moderate resistance (score 5) while ten families showed susceptibility (score 7) for sheath blight where as for bacterial leaf blight, five families recorded moderate resistance (score 5) while eighteen families showed susceptibility (score 7) (Table II).

TABLE II: Screening of F_5 families for sheath blight and bacterial leaf blight resistance

S.No	Cross	Number of families screened	Number of families with score		
			5	7	9
	Sheath blight resistance				
1	MTU 7029/ NLR 34449	12	5	7	-
2	MTU 7029/ PAU 3116-25-5-1	23	13	10	-
	Total	35	18	17	-
	Bacterial leaf blight resistance				
1	MTU 7029/ NLR 34449	12	2	10	-
2	MTU 7029/ PAU 3116-25-5-1	23	5	18	-
	Total	35	7	28	-

In total, out of 35 F5 families, 18 families reported moderate resistance to sheath blight while 17 families were susceptible. Regarding bacterial leaf blight, only seven families showed moderate resistance while 28 families were susceptible. Diseases resistance scores for all the 35 F5 families were provided in table 4. Graphical representation for number of families under each cross was represented in Fig 1. Similar results were reported by Yadav et al. (2015) for sheath blight and Ahmed Khan et al. (2009) and Thimmegowda et al. (2011) for bacterial leaf blight.

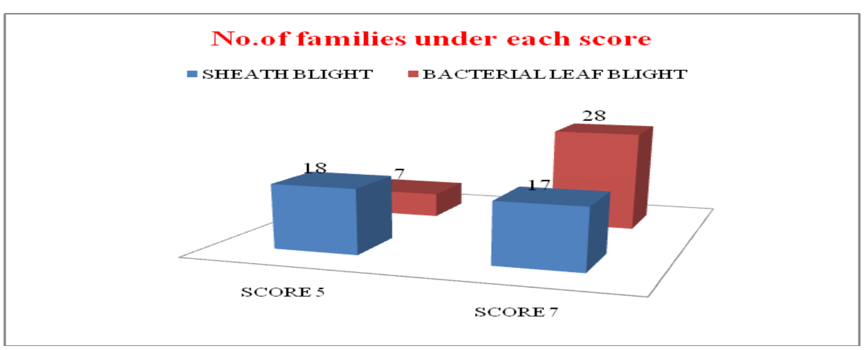

Fig. 1: Graphical Representation of Number of Families of Each Cross under each Score

Five families MTU 2465-24-3-1, MTU 2465-9-3-1, MTU 2468-7-2-1, MTU 2468-25-1-2 and MTU 2468-20-2-1 recorded moderate resistance to both the diseases and hence can be selected. (Table III).

TABLE III: Screening scores of 35 F_5 families

S.No	Entry	Cross Combination	Scores		Remarks
			Sheath blight	Bacterial leaf blight	
1	MTU 2465-16-2-1	MTU 7029/NLR 34449	7	7	Susceptible to both the diseases
2	MTU 2465-27-1-1	MTU 7029/NLR 34449	5	7	Moderately resistant to sheath blight but susceptible to bacterial leaf blight
3	MTU 2465-11-2-1	MTU 7029/NLR 34449	5	7	Moderately resistant to sheath blight but susceptible to bacterial leaf blight
4	MTU 2465-16-1-1	MTU 7029/NLR 34449	7	7	Susceptible to both the diseases
5	MTU 2465-35-2-1	MTU 7029/NLR 34449	7	7	Susceptible to both the diseases
6	MTU 2465-23-1-1	MTU 7029/NLR 34449	7	7	Susceptible to both the diseases
7	MTU 2465-3-1-1	MTU 7029/NLR 34449	7	7	Susceptible to both the diseases
8	MTU 2465-35-1-1	MTU 7029/NLR 34449	5	7	Moderately resistant to sheath blight but susceptible to bacterial leaf blight
9	MTU 2465-22-3-1	MTU 7029/NLR 34449	7	7	Susceptible to both the diseases
10	MTU 2465-12-2-1	MTU 7029/NLR 34449	7	7	Susceptible to both the diseases
11	MTU 2465-24-3-1	MTU 7029/NLR 34449	5	5	Moderately resistant to both the diseases
12	MTU 2465-9-3-1	MTU 7029/NLR 34449	5	5	Moderately resistant to both the diseases
13	MTU 2468-1-2-1	MTU 7029/ PAU 3116-25-5-1	5	7	Moderately resistant to sheath blight but susceptible to bacterial leaf blight
14	MTU 2468-31-1-2	MTU 7029/ PAU 3116-25-5-1	5	7	Moderately resistant to sheath blight but susceptible to bacterial leaf blight
15	MTU 2468-6-1-1	MTU 7029/ PAU 3116-25-5-1	7	7	Susceptible to both the diseases
16	MTU 2468-30-2-1	MTU 7029/ PAU 3116-25-5-1	5	7	Moderately resistant to sheath blight but susceptible to bacterial leaf blight
17	MTU 2468-7-2-1	MTU 7029/ PAU 3116-25-5-1	5	5	Moderately resistant to both the diseases
18	MTU 2468-25-2-2	MTU 7029/ PAU 3116-25-5-1	5	7	Moderately resistant to sheath blight but susceptible to bacterial leaf blight
19	MTU 2468-5-1-2	MTU 7029/ PAU 3116-25-5-1	5	7	Moderately resistant to sheath blight but susceptible to bacterial leaf blight
20	MTU 2468-20-1-3	MTU 7029/ PAU 3116-25-5-1	7	7	Susceptible to both the diseases
21	MTU 2468-25-3-1	MTU 7029/ PAU 3116-25-5-1	7	7	Susceptible to both the diseases
22	MTU 2468-5-2-1	MTU 7029/ PAU 3116-25-5-1	5	7	Moderately resistant to sheath blight but susceptible to bacterial leaf blight
23	MTU 2468-27-1-2	MTU 7029/ PAU 3116-25-5-1	7	7	Susceptible to both the diseases
24	MTU 2468-12-1-1	MTU 7029/ PAU 3116-25-5-1	7	7	Susceptible to both the diseases
25	MTU 2468-25-1-2	MTU 7029/ PAU 3116-25-5-1	5	5	Moderately resistant to both the diseases
26	]MTU 2468-32-1-1	MTU 7029/ PAU 3116-25-5-1	5	7	Moderately resistant to sheath blight but susceptible to bacterial leaf blight
27	MTU 2468-25-1-1	MTU 7029/ PAU 3116-25-5-1	5	7	Moderately resistant to sheath blight but susceptible to bacterial leaf blight
28	MTU 2468-6-1-2	MTU 7029/ PAU 3116-25-5-1	7	7	Susceptible to both the diseases
29	MTU 2468-5-1-1	MTU 7029/ PAU 3116-25-5-1	7	7	Susceptible to both the diseases
30	MTU 2468-20-1-2	MTU 7029/ PAU 3116-25-5-1	7	5	Moderately resistant to bacterial leaf blight but susceptible to sheath blight
31	MTU 2468-32-1-2	MTU 7029/ PAU 3116-25-5-1	5	7	Moderately resistant to sheath blight but susceptible to bacterial leaf blight
32	MTU 2468-20-2-1	MTU 7029/ PAU 3116-25-5-1	5	5	Moderately resistant to both the diseases
33	MTU 2468-27-1-1	MTU 7029/ PAU 3116-25-5-1	7	5	Moderately resistant to bacterial leaf blight but susceptible to sheath blight
34	MTU 2468-10-1-1	MTU 7029/ PAU 3116-25-5-1	5	7	Moderately resistant to sheath blight but susceptible to bacterial leaf blight
35	MTU 2468-5-2-2	MTU 7029/ PAU 3116-25-5-1	7	7	Susceptible to both the diseases
36	MTU 7029		9	-	Highly susceptible check for sheath blight
37	TN-1		-	9	Highly susceptible check for bacterial leaf blight

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