



International Journal of Applied Sciences and Biotechnology

A Rapid Publishing Journal

ISSN 2091-2609

Indexing and Abstracting

CrossRef, Google Scholar, Global Impact Factor, Genamics, Index Copernicus, Directory of Open Access Journals, WorldCat, Electronic Journals Library (EZB), Universitätsbibliothek Leipzig, Hamburg University, UTS (University of Technology, Sydney): Library, International Society of Universal Research in Sciences (EyeSource), Journal Seeker, WZB, Socolar, BioRes, Indian Science, Jadoun Science, Jour-Informatics, Journal Directory, JournalTOCs, Academic Journals Database, Journal Quality Evaluation Report, PDOAJ, Science Central, Journal Impact Factor, NewJour, Open Science Directory, Directory of Research Journals Indexing, Open Access Library, International Impact Factor Services, SciSeek, Cabell's Directories, Scientific Indexing Services, CiteFactor, UniSA Library, InfoBase Index, Infomine, Getinfo, Open Academic Journals Index, HINARI, etc.

CODEN (Chemical Abstract Services, USA): IJASKD

Vol-3(1) March, 2015

Available online at:

http://www.ijasbt.org & http://www.nepjol.info/index.php/IJASBT/index



Impact factor*: **1.422** Scientific Journal Impact factor#: **3.419** Index Copernicus Value: **6.02**

*Impact factor is issued by Universal Impact Factor. Kindly note that this is not the IF of Journal Citation Report (JCR). #Impact factor is issued by SJIF INNO SPACE.



Research Article

SCREENING OF RICE VARIETIES AGAINST BROWN LEAF SPOT DISEASE AT JYOTINAGAR, CHITWAN, NEPAL

Prem Bahadur Magar^{*}

Karma & Sons Traders Pvt. Ltd., Kathmandu, Nepal

*Corresponding author's email: premmagar12@gmail.com

Abstract

A field experiment was conducted on fourteen rice varieties to determine the level of resistance against brown leaf spot (*Bipolaris oryzae*) disease at Karma Research and Development Center, Jyotinagar, Chitwan during summer in 2013. Design used was randomized complete block design with three replications. Disease severity, total AUDPC value, thousand grain weight and grain yield were highly significant among the rice varieties. Disease severity and total AUDPC value ranged from 21.73% to 58.07% and 614.8 to 1827 respectively. None of these varieties were found either resistant or highly resistant to disease. Only HJ-G1 and HJ-G2 were moderately resistant with grain yield of 5.10 t/ha and 4.25 t/ha respectively. HJ-G1 variety had the highest grain yield 5.10 t/ha and lowest disease severity (21.73%) as well as total AUDPC value (6148). From this experiment, it can be concluded that HJ-G1 variety can be used for higher grain yield purpose under similar field condition because it has highest yield and also tolerant to disease.

Key words: Resistance; Brown leaf spot; Bipolaris oryzae; Disease severity; AUDPC; Grain yield.

Introduction

Rice (*Oryza sativa* L.) is the world's single most important food crop and a primary food for more than a third of the world's population (Khush and Toenniessen, 1991). Globally, more than 3.5 billion people depend on rice for more than 20% of their daily calories (IRRI, 2012). In Nepal, rice is grown in 1.53 million ha land with total production of 5.07 million Mt and yield of 3,312 kg per hectare (MoAD, 2012). The terai region (60-900 masl) contains 69.73% of the total rice area and contributes 73.24% of total rice production. Hills (900-1500 masl) and mountains (1500-2750 masl) have 25.82% and 4.44% of total rice area producing 23.71% and 3% of rice production (MOAD, 2012). Rice is the 1st staple food grain crop of Nepal and has a significant role in food security of the Nepalese people.

Because of its devastating nature, widespread distribution and existence of several physiological races of the pathogen (*Bipolaris oryzae*), brown leaf spot disease is the most serious disease of rice (Arshad *et al.*, 2008). It caused Bengal Famine in 1942, with yield loss of 50-90%, which resulted in death of 2 million people due to starvation. The pathogen can infects both seedlings and mature plants with the coleoptile, leaves, leaf sheath, panicle branches, glumes, and spikelets (Webster and Gunnell, 1992). The disease is also known as poor rice farmer's disease because it occurs mostly in deficient and poor soils (Agarwal, 1989; Mia, 1998; Zadoks, 2002). The disease has been noted to reduce yields from 6 to 90% in Asia (Padmanabhan, 1973; Estrada, 1984; Mew and Gonzales, 2002).

Although fungicide treatment is available and low cost for the management of brown leaf spot, host plant resistance is most economical. Similarly, fungicides use becomes even less favorable in the future as a result of restrictions in their use (Hovmoller, 2001). Use of resistant varieties is a simple, effective, safe and economical means of controlling rice diseases. Resistant varieties help to stabilize yield. The resistance is very scarce and not stable due to the appearance of new or more virulent races of pathogens (Katasntonis *et al.*, 2007). So, resistant level should be updated each year for each variety. In such contest, identification of resistant genotypes or varieties would be good alternatives to manage brown leaf spot.

Material and Methods

The field experiment was conducted at Karma Research and Development Center (KRDC), Jyotinagar, Chitwan, Nepal during summer (June to November, 2013). The site lies in the sub-tropical zone with an altitude of 250 masl, between $27^{\circ}36'$ N latitude and $84^{\circ}16'$ E longitude. Design used for the experiment was randomized complete block design (RCBD) with 3 replications. Each replication was separated by 1 m and there was 0.5 m gap between each individual plot within the replication. Fourteen treatments were laid in one replication as seven in each line. Individual plot size was 9 m² (3 m × 3 m) and total area of the research field was 611 m^2 . There were 15 rows of 3 m length/plot and 20 cm apart. Plant to plant spacing was 20 cm with single seedlings per hill for all varieties.

Seeds of eleven rice varieties were collected from Hi-tech Seed Co. Ltd., China along with popular rice hybrids in Nepal, DY-18 and DY-69, and improved variety OR (Ramdhan) was used in the study. Transplanting was done manually with one seedling (29 days old) per hill at 16th July, 2013 and gap filling was done 7 days after transplanting to maintain desired plant population. The dose of chemical fertilizer applied was 100:30:30 kg NPK per hectare through urea (46% N), DAP (18% N and 46% P₂O₅) and MOP (60% K₂O). Half dose of Nitrogen, full dose of Phosphorus and Potash were applied before final land preparation as basal dose. Remaining dose of N was applied in two split doses at active tillering stage and panicle initiation. Zinc Sulphate (commercial product) was applied @ 20 kg/ha at final land preparation. Irrigation was given in the field as per requirement and about 5 cm height of water was maintained up to the grain filling stage of rice. Twice spraying of Kingstar (emamactin benzoate 5% SG) @ 5 g/161 of water and Kingvan (dichlorovos 80% EC) @ 2 ml/l of water was applied before milking stage to control rice gundhi bug and stem borer.

Disease assessment

Randomly selected 25 plants were tagged from each plot for disease scoring. Disease was recorded from all sample plants. Starting with the appearance of the first brown leaf spot disease symptoms, tagged plants within each plot was visually evaluated for percent foliar infection (severity) at fifteen days interval. A total of 4 scorings were done from August to October, 2013, i.e. August 21, September 5, September 20 and October 5. The effect of disease (severity) on rice variety was integrated into area under disease progress curve (AUDPC), as described by Campbell and Madden (1990). Disease scoring was done by using standard disease rating scale of IRRI (2002) (Table 1).

Percentage disease intensity was calculated using the following formula:

Disease intensity % =
$$\frac{\text{Sum of all numerical ratings} \times 100}{\text{No. of plants observed} \times \text{maximum rating}}$$

Area under disease progress curve (AUDPC) gives a quantitative measure of disease development and intensity of disease (Reynolds and Neher, 1997), and it helps to categorize varieties under different level of resistance. It also summarizes the progress of disease severity along a time period and was estimated using the following formula as given by Campbell and Madden (1990).

$$\begin{array}{l} n\text{-}1 \\ AUDPC = \sum\limits_{i=1}^{n-1} (Y_{i+1} + Y_i) \ 0.5 \ (T_{i+1} - T_i) \end{array}$$

Where,

- Y_i = brown leaf spot disease severity on the ith date
- $T_i = date on which the disease was scored$

n = numbers of dates on which disease was scored

Table 1:	Disease rating scale used for screening of rice
	varieties against brown spot caused by Bipolaris
	010170700

Scale	Affected leaf area	Host response
1	No incidence	Immune
2	Less than 1%	Highly Resistant
3	1 – 3%	Resistant
4	4-5%	Resistant
5	11 – 15%	Moderately Resistant
6	16-25%	Moderately Resistant
7	26-50%	Susceptible
8	51 - 75%	Susceptible
9	76 - 100%	Highly susceptible

Agronomic traits

At maturity i.e. November 7, 2013, plants were harvested from each plot, threshed manually; grain weights and thousand grain weights were taken using digital balance. Then, grain yield was converted into ton per hectare. Digital moisture meter was used to record the moisture percentage of the grain at the time of weighing. Finally grain yield was adjusted at 12% moisture level using the formula.

Gain yield (t/ha) at 12% moisture = $\frac{(100 - MC) \times Plot yield (kg) \times 10}{(100 - 12) \times net plot area (m2)}$

Statistical analysis

Data entry and processing was carried out using MS-excel 2007 program. The data were processed to fit into MSTAT-C (Freed and Scott, 1986) software for analysis. DMRT was done at 1% and 5% level of significance for mean comparison from the reference of Gomez and Gomez (1984) and was applied to identify the most resistant varieties.

Results and Discussion

Effect of disease severity and AUDPC in rice varieties

Symptoms of brown leaf spot disease appeared in all rice varieties. The result showed that there was highly significant difference among rice varieties in terms of disease severity and total AUDPC value ($p \le 0.01$) (Table 2).

Disease severity varied considerably among rice varieties which ranged from 21.73% to 58.07% (Table 3). Highest severity was found on HJ-G7 (58.07%) while HJ-G1 (21.73%) had significantly lowest disease severity. Also, there was significant difference in area under disease progress curve (AUDPC) values among them in all 3 observation dates. AUDPC values increased with time of observation in all rice varieties. The variety HJ-G1 had the lowest and HJ-G7 had the highest AUDPC values in all observation dates, with a total AUDPC value of 614.8 and

1827, respectively (Table 3). The variation in disease increment might be due to variation in susceptibility of varieties to the pathogen.

Response of rice varieties to brown leaf spot disease

Rice varieties screened against brown leaf spot disease showed different response during summer at KRDC, Jyotinagar, Chitwan. None of these varieties were found resistant on the basis of standard disease rating scale given by IRRI. Most of them belong to susceptible category. The resistance level of different varieties against the disease is shown in the Table 4.

Use of resistant variety is the cheapest and ideal method of controlling the disease. For managing the brown leaf spot disease, the most desirable means is host resistance, especially in developing countries (Bonman, 1992). Sato *et al*, 2008 reported three QTLs against brown spot. In this study; among the 14 varieties, only HJ-G1 and HJ-G2 were found as moderately resistant and rests of them were susceptible to disease. In Pakistan, Arshad *et al.*, 2008 found only one entry among seventy entries was resistant against brown leaf spot. Variability in rice germplasm in response to various diseases was also reported by Hossain and Kulkarni (2001) and Castano *et al.* (1990). They also categorized rice germplasm into different groups ranging from highly susceptible to highly resistant against various rice diseases. Saifullah *et al.* (1991) and Nagaraju *et al.* (1991) have also showed significant variability in rice genotypes against diseases.

Table 2: Mean square value of brown leaf spot disease severity, total area under disease progress curve (AUDPC), thousand grain weight and grain yield on 14 rice varieties during 2013 cropping season at Jyotinagar, Chitwan.

Source	Df	Disease severity (%)	Total AUDPC value	Thousand grain weight (g)	Grain yield(t/ha)
Replication	2	0.173 ^{ns}	395.510 ^{ns}	1.152 ^{ns}	0.305 ^{ns}
Varieties	13	400.056**	500579.824**	13.673**	2.199**
Error	26	18.361	7051.275	1.732	0.476
Total	41				

Df: degree of freedom, ns: statistically non significant, * significant at 0.05 level of significance, ** significant at 0.01 level of significance.

		Disease severity (%) (80 DAT)	AUDPC Values					Grain yield
S.N.	Variety		5-Sep (50 DAT)	20-Sep (65 DAT)	5-Oct (80 DAT)	Total AUDPC Value	TGW (g)	(t/ha)
1	HJ-G1	21.73 ^g	117.8 ^g	204.5 ^f	292.5 ^f	614.8 ^f	33.09 ^a	5.10 ^a
2	HJ-G2	24.30 ^{fg}	223.5 ^{de}	383.8 ^c	537.0 ^{bc}	1144.0 ^d	30.40 ^{bc}	4.25 ^{abcd}
3	HJ-G3	43.17 ^{cd}	233.0 ^d	378.0 ^c	475.0 ^{cd}	1086.0 ^d	28.91 ^{cd}	3.17 ^{de}
4	HJ-G5	53.73ª	424.5 ^{ab}	613.3ª	745.3ª	1803 ^{ab}	27.09 ^d	2.83 ^e
5	HJ-G7	58.07 ^a	456.0ª	625.8ª	765.0 ^a	1827 ^a	29.87°	2.70 ^e
6	HJ-G8	46.10 ^{bc}	230.0 ^d	358.0 ^{cd}	539.5 ^{bc}	1128.0 ^d	30.11b ^c	3.37 ^{cde}
7	HJ-G9	33.73 ^e	188.8 ^{def}	273.0 ^e	327.8 ^{ef}	789.5 ^e	28.16 ^{cd}	4.80 ^a
8	HJ-G10	54.67ª	391.1 ^b	600.6 ^a	814.0 ^a	1806 ^{ab}	29.13 ^{cd}	2.76 ^e
9	HJ-G11	42.77 ^{cd}	381.3 ^b	567.8 ^a	721.8 ^a	1671.0 ^b	30.68 ^{bc}	2.99 ^{de}
10	HJ-G12	52.50 ^{ab}	302.3°	469.5 ^b	611.0 ^b	1383.0 ^c	32.45 ^{ab}	2.91 ^e
11	HJ-G13	32.43 ^e	153.5 ^{fg}	307.8 ^{de}	462.8 ^{cd}	924.0 ^e	30.60 ^{bc}	3.17 ^{de}
12	DY-18	35.87 ^{de}	170.5 ^{efg}	326.4 ^{cde}	441.7 ^d	938.6 ^e	29.32 ^{cd}	4.47 ^{abc}
13	DY-69	42.40 ^{cd}	206.0 ^{def}	291.0 ^{de}	397.5 ^{de}	894.5 ^e	29.13 ^{cd}	4.73 ^{ab}
14	OR	29.90 ^{ef}	323.0 ^c	491.3 ^b	606.8 ^b	1421.0 ^c	24.45 ^e	3.51 ^{bcde}
SEm ((±)	2.474	17.95	21.47	29.35	48.48	0.7598	0.3983
CV (%	%)	10.50	11.45	8.84	9.20	6.75	4.46	19.04
LSD ₀ .	.05 value	7.192	52.19	62.41	85.3	140.9	2.209	1.158

 Table 3: Mean value of brown leaf spot disease severity, area under disease progress curve (AUDPC), total AUDPC, thousand grain weight (TGW) and grain yield of 14 rice varieties at Jyotinagar, Chitwan.

DAT: Days after transplanting, means in a column followed by the same letters are not significantly different according to LSD at 5% probability level, CV: Coefficient of variance and LSD_{0.05}: Least Significant Difference at 5% level of significance, SEm (\pm) indicates standard error of mean

Table 4: Resistance category of 14 rice varieties to brown
leaf spot disease on the basis of disease severity.

S.N.	Variety	Resistance category
1	HJ-G1	MR
2	HJ-G2	MR
3	HJ-G3	S
4	HJ-G5	S
5	HJ-G7	S
6	HJ-G8	S
7	HJ-G9	S
8	HJ-G10	S
9	HJ-G11	S
10	HJ-G12	S
11	HJ-G13	S
12	DY-18	S
13	DY-69	S
14	OR	S

MR: Moderately resistant, S: Susceptible

Yield and thousand grain weight

ANOVA (Analysis of variance) showed, there was highly significant difference in grain yield and thousand grain weight (TGW) among rice varieties ($p \le 0.01$) (Table 2). Highest grain yield (5.10 t/ha) was found in HJ-G1 with total AUDPC value of 614.8 while lowest (2.70 t/ha) in HJ-G7 with 1827, total AUDPC value (Table 3). However, grain yield of HJ-G1 variety didn't differ significantly with HJ-G9 (4.80 t/ha), DY-69 (4.73 t/ha), DY-18 (4.47 t/ha) and HJ-G2 (4.25 t/ha). Yield potential differed among the varieties and it might be influenced by the level of disease. HJ-G1 variety had 1.59 t/ha greater yield than local improved variety OR.

Thousand grain weights (TGW) were highly significant among the rice varieties. The TGW ranged from 24.45 g to 33.09 g. Highest TGW 33.09 g was found in HJ-G1 and lowest in OR (24.45 g). From the result, it was found that the variety, HJ-G7, showing the maximum disease severity (58.07%), also showed the highest total AUDPC value (1827) and least yield (2.70 t/ha). So, this genotype appeared as the most susceptible variety to brown leaf spot among all the tested varieties in field under natural infection condition. Similarly, the variety HJ-G1 showed the maximum yield (5.10 t/ha) which has the lowest disease severity (21.73%) and also the least, total AUDPC value (614.80). Thus, the variety HJ-G1 was suitable for higher grain yield in summer at Chitwan condition than other varieties.

Conclusion

The screening of fourteen rice varieties against brown leaf spot disease revealed that none of the varieties was immune. Among them, only HJ-G1 and HJ-G2 were found moderately resistant. Highest grain yield (5.10 t/ha) was found in HJ-G1 with least disease severity of 21.73%. Also, HJ-G1 had the lowest AUDPC values in all observation dates, with a total AUDPC value of 614.8. So, it is recommended to use HJ-G1 variety because it has highest yield in comparison to other varieties as well as tolerant to disease.

Acknowledgement

I would like to acknowledge Hi-tech Seed Co. Ltd., China for providing the seed sample. Thanks to Mr. Suresh Gurung, M. D. of Karma & Sons Traders Pvt. Ltd., for financial support, Research and Development Managers Mr. Amit Raj Adhikari (Soil Scientist) and Mr. Bishnu Pandey (Agronomist), Marketing Executive Mr. Basanta Rajbanshi, and other staffs at KRDC, Jyotinagar, Chitwan for their support to finally accomplish this research.

References

- Agarwal VK (1989) A simpler technique for routine examination of rice seed lots for rice brown leaf spot disease. Seed Technology News. Bulletin of the Indian society of seed technology. **11**: 1-2.
- Arshad HMI, Khan JA and Jamil FF (2008) Screening of rice germplasm against blast and brown spot diseases. *Pak. J. Phytopathol.* 20(1): 52-57.
- Bonman JM (1992) Durable resistance to rice brown leaf spot disease environmental influences. *Euphytica*. 63 (2): 115-123. DOI: 10.1007/BF00023917
- Campell CL and Madden LV (1990) Introduction to plant disease Epidemology. John Wiley and Sons, New York. 532 p.
- Castano J, Amril B, Syahril D and Zaini Z (1990) Upland rice genotypes resistant to blast disease in west Sumatra. *Int. Rice Res. Newslett.* 15: 11-12.
- Estrada AB (1984) Selection of differential varieties for race study of *Helminthosporium oryzae*. M.S. thesis. College, Leguna, the Philippines: University of Philippines at Las Banos, Philippines.
- Freed RD and Scott DE (1986) MSTATC. Crop and soil. Michigan: Michigan State University, USA.
- Gomez KA and Gomez AA (1984) Statistical procedures for agricultural research (2nd ed). A Wiley Interscience Publication, New York, USA. 655 p.
- Hossain MM and Kulkarni S (2001) Field evaluation of fungicides, neem based formulation and biological agents against blast of rice. J. Maharashtra. Agric. Uni. 26: 148-150.
- Hovmoller MS (2001) Disease severity and pathotype dynamics of *Puccinia striiformis* f. sp. *tritici* in Denmark. *Plant pathology*. **50**: 181-189. DOI: 10.1046/j.1365-3059.2001.00525.x
- IRRI (2002) Standard Evaluation System for Rice (SES). International Rice Research Institute. Manila, Philippines. 56 p.
- IRRI (2012) Rice facts. International Rice Research Institute. Manila, Philippines.

Katasntones G, Koutrovbas SD, Ntanos DA and Lupoho E (2007) A comparison of three experimental designs for the field assessment of resistance to rice brown leaf spot of rice disease (*Bipolaris oryzae*). *J. Phytopath.* **155**: 204-210. DOI: 10.1111/j.1439-0434.2007.01218.x

- Khush GS and Toenniessen GH (1991) Rice Biotechnology. International Rice Research Institute (IRRI) and CAB International. 258 p.
- Mew TW and Gonzales P (2002) A handbook of seed borne fungi. Las Banos, Laguna, Philippines. pp. 17-20.
- Mia MAT (1998) Brown spot of rice. 1st national workshop on seed pathology, 9-12 June 1998. Organized by Danish Govt. Institute of seed pathology and seed pathology and seed pathology laboratory. BAU, Mymensingh.
- MoAD (2012) Statistical information on Nepalese agriculture. Government of Nepal. Ministry of Agricultural Development. Agribusiness Promotion and Statistics Division, Singha Durbar, Kathmandu, Nepal. 179 p.
- Nagaraju Vasanthakumar HC, Devaiah BM, Sehadri VS and Naidu BS (1991) Evaluation of rice genotypes for blast and sheath rot resistance in hilly region of Karnataka. *Mysore J. Agric. Sci.* **25**: 139-141.

- Padmanabhan SY (1973) The Great Bengal famine. Ann. rev.Phytopathology.11:11-26.DOI:10.1146/annurev.py.11.090173.000303
- Reynolds L and Neher DA (1997) Statistical comparison of epidemics. *In:* L. J. Francl and D. A. Neher (eds.), Exercises in Plant Disease Epidemiology. APS Press, St. Paul, USA. pp. 34-37.
- Saifulla M, Nagaraju Vasanthakumar ML and Devaiah BM (1991) Evaluation of promising rice genotypes for blast resistance. Oryza 28: 281-282.
- Sato H, Ando I, Hirabayashi H, Takeuchi Y, Arase S, Kihara J, Kato H, Imbe T, and Nemoto H (2008) QTL analysis of brown spot resistance in rice (*Oryza sativa* L.). *Breed. Sci.* 58: 93-96. DOI: 10.1270/jsbbs.58.93
- Webster RK and Gunnell PS (1992) Compendium of rice diseases. American Phytopathological Society, St. Paul, Minnesota. 62 p.
- Zadoks JC (2002) Fifty years of crop protection, 1950-2000. *Neth. J. Agric. Sci.* **2002**: 181-193.