International Journal of Pharmacy and Biological Sciences-IJPBS™ (2019) 9 (2): 131-137
Online ISSN: 2230-7605, Print ISSN: 2321-3272
Research Article | Biological Sciences | Open Access | MCI Approved
UGC Approved Journal

Integrated Rice Brown Leaf Spot Management Under Pot Culture Conditions

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Received: 18 Jan 2019 / Accepted: 16 Mar 2019 / Published online: 1 Apr 2019
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Abstract
The Pot culture studies were undertaken to investigate the integrated disease management in rice as influenced by application of Zinc sulphate and foliar application of salicylic acid and Potassium silicate and Brown spot pathogen *Bipolaris oryzae* inoculation. The results revealed that Soil application of Zinc sulphate @ 25 kg/ ha along with foliar application of plant activator Salicylic acid @ 50 ppm on 15 days after transplanting and Foliar spray of silicon based nutrient potassium silicate @ 3 % recorded the minimum disease incidence and maximum biometrics of rice. Also, the results revealed that the same treatment recorded maximum plant height, number of tillers per clump, number of productive tillers per clump, panicle length, thousand grain weight, filler grain percentage, grain yield and straw yield when compared to control and comparison fungicide treatments.

Keywords
Integrated disease management, brown spot, Salicylic acid, Potassium silicate, macro-micro nutrient.

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INTRODUCTION
Rice, which is being cultivated for several years in our country, it is not just a grain, it is the lifeline and is the second most important crop next to wheat. India is one among the leading producer of rice in Asia (Tony Cisse, 2005). Rice crop has been under cultivation from time immemorial, being grown under varying climatic conditions in different parts of the country. It is widely affected by quite a number of diseases caused by fungi, bacteria, viruses and mycoplasma which results in higher yield losses (Ou, 1985). Among the various fungal diseases of rice, brown spot or sesame leaf spot incited by *Helminthosporium oryzae* (Breda de Haan) Subram and Jain (Current name: *Bipolaris oryzae* (Breda de Haan) Shoemaker) is found to occur in most rice growing areas.

Currently the disease is being managed by application of fungicides. Due to pesticides hazards, pollution effect, fungicide resistant, bio control agent resistant strains, lack of bio protectant knowledge which required the integrated component approach in Indian farmer's level which will be improve growth and disease suppression.

Mineral nutrition has long been recognised as an important component of disease control practices (Huber and Wilhelm, 1988). Mineral nutrients can increase or decrease in the development of diseases caused by different organisms. The mechanisms leading to these nutrients induced changes in disease development are complex, multifarious, includes

effects of mineral nutrients directly on the pathogen, on plant growth development and on plant resistance mechanisms (Walters and Bingham, 2007). A balanced fertilization program with macro-micronutrients in plant nutrition is very important in the production of high yield with high quality crop produces (Sawan *et al.,* 2001; Nasiri *et al.,* 2010).

Therefore, with an aim to develop an integrated strategy involving the use of certain macro-micro nutrients, silicon based nutrients and resistance inducing chemicals for the successful sustainable management of rice brown spot. Hence, the present studies were undertaken to investigate the nutrient analysis of macro nutrient content of rice by application of Macro-micro nutrient, Salicylic acid, potassium silicate along with pathogen inoculation.

MATERIALS AND METHODS

Crop, Variety and Source

Crop : Rice (*Oryza sativa* L.)
Variety : ADT 36
Source : Tamil Nadu Rice Research Institute (TRRI), Aduthurai, Tamil Nadu.

-Pot culture studies

The pot culture studies were conducted to test the efficacy of certain macro-micro nutrients, silicon based nutrients and certain resistance inducing chemicals for assessing their influence on the incidence of brown spot of rice with various treatment and combinations. The brown spot susceptible variety ADT 36 grown in rectangular pots of size, 30x45 cm was used for the study. The plants were given artificial inoculation by spraying the spore suspensions with adequate spore load (50,000 spores/ml) at 15 DAT in the evening hours. The crop was maintained in a poly house with frequent spraying of water to provide adequate moisture and relative humidity to enable successful infection by the pathogen. The experiments were conducted in a randomized block design with three replications for each treatment and a suitable control. The fungicide carbendazim 50 WP @ 0.1 per cent was used for comparison and the standard agronomic practices as recommended by the State Agricultural Department were followed.

The effective treatments observed in different experiments conducted under pot and field conditions were pooled together and a new schedule of treatments in combination was evolved for the effective management of brown spot disease of rice. Also, zinc sulphate @ 25 Kg/ha was applied as basal application to the entire treatments (ZSS) except control and comparison. The treatment details are given below;

Treatment schedule

T_1 – ZSS + ZSF_1 + ZSF_2
T_2 – ZSS + SA_1 + SA_2
T_3 – ZSS + PS_1 + PS_2
T_4 – ZSS + ZSF_1 + SA_2
T_5 – ZSS + SA_1 + ZSF_2
T_6 – ZSS + SA_1 + PS_2
T_7 – ZSS + PS_1 + SA_2
T_8 – ZSS + PS_1 + ZSF_2
T_9 – ZSS + ZSF_1 + PS_2
T_{10} – Carbendazim 50 WP @ 0.1 per cent as foliar spray (comparison)
T_{11} – Control $ZnSO_4$ @ 25 Kg/ha was applied as basal application to the entire treatments (ZSS) except control and comparison. The treatment details are given below;

T_1 – ZSS + Two sprays of zinc sulphate @ 3 % on 15 and 30 DAT
T_2 - ZSS + Two sprays with salicylic acid @ 50 ppm on 15 and 30 DAT.
T_3 - ZSS + Two sprays with potassium silicate @ 3 % on 15 and 30 DAT.
T_4 - ZSS + First spray with zinc sulphate @ 3 % on 15 DAT + second spray with salicylic acid @ 50 ppm on 30 DAT.
T_5 - ZSS + Second spray with zinc sulphate @ 3 % on 30 DAT
T_6 - ZSS + First spray with salicylic acid @ 50 ppm on 15 DAT + second spray with potassium silicate @ 3 % on 30 DAT
T_7 - ZSS + First spray with potassium silicate @ 3 % on 15 DAT + second spray with salicylic acid @ 50 ppm on 30 DAT
T_8 - ZSS +First spray with potassium silicate @ 3 % on 15 DAT + second spray with zinc sulphate @ 3 % on 30 DAT
T_9 - ZSS + First spray with zinc sulphate @ 3 % on 15 DAT + second spray with potassium silicate @ 3 % on 30 DAT
T_{10} – Carbendazim (0.1 %) – Comparison
T_{11} - Un treated control.

Disease assessment

In all the studies observations on disease incidence, grain and straw yield were recorded at the time of harvest. The disease incidence was observed from a randomly selected set of three hills per pot.

Grain yield

After the harvest, the grains were separated, winnowed, dried in the sun and dry weight was recorded and expressed as g/pot.

Straw yield

After thrashing and separation of grains, the straw was dried pot wise in sun for two days. Later the straw weight was recorded and expressed as g/pot.

Plant height

The height of the plant was measured from the surface of the soil to the neck of panicle. Plant height was measured at maturity stage on sample plants and the mean height was calculated and recorded in cm.

Number of tillers per clump

Number of tillers per clump was recorded at maximum tillering stage by taking average of randomly chosen five clumps.

Number of productive tillers per clump

The productive tillers per clump were counted from total number of tillers at maturity stage in the five sample plants and the mean value was calculated and recorded.

No. of grains per panicle

The length of the panicle was measured after the harvest and the mean length was calculated and recorded in cm.

Thousand grain weight

Thousand filled grains were counted from the bulk of grains drawn at random in each plot dried to 14 per cent moisture, weighed and expressed in gram (Yoshida *et al.,* 1972).

The disease incidence was assessed by adopting 0-9 scale according to "Phytopathometry" by Mayee and Datar (1986) and the per cent disease incidence /index was calculated based on the formula suggested by Vidhyasekaran *et al.* (1989).

DISEASE SEVERITY	DESCRIPTION OF DISEASE INDEX
0	No lesions
1	Affected leaf area less than 1 %
3	1-10 % affected leaf area
5	11-25 % affected leaf area
7	26 -50 % affected leaf area
9	> 50 % leaf area affected

$$\textbf{Per cent Disease Index} = \frac{\text{Total ratings}}{\text{Total number of leaves graded} \times \text{Maximum grade in the score chart.}} \times 100$$

Statistical analysis

The statistical analysis of the experimental results was performed employing the computer software package 'IRRISTAT', version 90-1, developed by Department of Statistics, International Rice Research Institute, Philippines and as per the procedure of Gomez and Gomez (1976).

RESULTS AND DISCUSSION

Effect of ZS, SA, PS and *H.oryzae* inoculation on biometrics, per cent disease incidence and yield parameters of rice var. ADT 36

In pot culture experiments, combined application of zinc sulphate, salicylic acid and potassium silicate were found to be significantly superior over test fungicide Carbendazim in respect of reducing disease intensity and increasing biometrics and yield parameters. Results of the experiment (Table 1 and 2) showed that soil application of $ZnSO_4$ (ZSS) along with foliar application of SA_1 (15 DAT) and PS_2 (30 DAT) increased the plant height, number of tillers, number of productive tillers per clump, panicle length, thousand grain weight, percentage of filled grain, grain yield and straw yield when compared to other treatments.

Per cent disease incidence

Results depicted in table 1 showed that soil application of $ZnSO_4$ (ZSS) along with foliar application of SA_1 (15 DAT) and PS_2 (30 DAT) effectively (T_6) controlled the brown spot disease incidence (10.51 %). It was followed by (T_7) combined application of ZSS plus foliar application of potassium silicate (at 15 DAT) and SA (at 30 DAT) which recorded a disease incidence of 10.82 per cent. The test fungicide Carbendazim 0.1 per cent recorded 15.47 per cent while control recorded the maximum disease incidence with 60.67 per cent.

Plant height

Generally, all the treatments significantly increased the plant height when compared to Carbendazim treatment and control. The data mentioned, on the effect of different treatments on the height of rice crop was recorded in table 2. Among the treatments, the maximum plant height (89.56 cm) was observed in combined application of macro-micro nutrients

and inducing chemical (T_6) and control recorded the minimum plant height (72.65 cm).

No. of productive tillers / clump

The pot trial revealed that all the treatments with ZS, SA and PS had significantly higher number of productive tillers per clump, when compared to control. However, the treatment T_6 recorded maximum number of productive tillers (13.16) followed by T_7 (13.05). Control recorded the minimum tillers (10.12) and the fungicide Carbendazim recorded 11.28 number of tillers. Also, the maximum panicle length was recorded in T_6 (17.99 cm) when compared to comparison fungicide (16.84 cm) and control (16.00 cm).

Thousand grain weight

The data mentioned on the effect of various treatments on the thousand grain weight was recorded in table 2. All the treatments with ZS, SA and PS had increased the thousand grain weight when compared to control. However, the treatment T_6 recorded the maximum thousand grain weight of 20.65 g whereas the untreated control recorded the minimum thousand grain weight of 20.50 g.

Grain and Straw yield

The positive influence of various treatments on the grain and straw yield of rice was well established (Table 2) in the present investigation. All the treatments with ZS, SA and PS recorded significant increase in grain and straw yield, when compared to test fungicide and control. Among them T_6 (ZSS + SA_1 + PS_2) recorded the maximum filled grain percentage (83.06 %), maximum grain yield (35.60 g/pot) and straw yield (88.27 g/pot). The fungicide treatment recorded a grain yield of 30.86 g/pot and straw yield of 79.42 g/pot.

Nutrient management is an integral component of efficient production agriculture. Nutrient sufficiency may provide a general form of disease resistance by maintaining a high level of inhibitory compounds in tissue or quick response to invasion by a pathogen (Huber and Haneklaus, 2007). The inorganic salts like silicates, bicarbonates, phosphates, chlorides and phosphites have properties that make them desirable for inclusion in Integrated Disease Management (IDM) programmes (Deliopoulos *et al.,* 2010). Their proposed advantages include low cost, favourable safety profile for humans and the environment and low mammalian toxicity (Olivier *et al.,* 1999). Silicon can reduce the impact of nutrient deficiency of phosphorus in crop plants (Epstein, 1994) and can reduce the toxicity effects caused by aluminium and iron (Marschner, 1995) and induce defense responses (Fawe *et al.,* 2001). Thus, silicon fertilizer could be an environmental friendly alternative to manage crop diseases (Cai *et al.,* 2008). Application of micro nutrients such as Mn, Zn, Cu and B can increase the Ca^{2+} cations and interact with SA and activate ISR (Reuveni and Reuveni, 1998; Dordas, 2008). Zinc is a component of dehydrogenases, peptides and many other enzymes and also regulates synthesis of auxins in plant (Huber, 1981) and thus may play important role in conferring resistance to host. The effect of the micronutrients on reducing the severity of foliar diseases can be attributed to the involvement in physiology and biochemistry of the plant as the micronutrients are involved in many processes that can affect the response of plants to pathogens (Marschner, 1995). Deleterious effects of zinc to pathogen may include direct inhibition of spore germination (Singh *et al.*, 1987) and direct toxic effect on pathogen (Dordas, 2008).

Minnatullah and Jha (2002) reported that spraying of $ZnSO_4$ resulted in the lowest brown spot disease severity, highest grain yield in rice. Khan *et al.* (2010) reported that micronutrient mixture (Zn and Mn) sprayed at different crop growth stages in wheat enhanced the yield and grain characters. In light of the above, the present observations made on $ZnSO_4$ spray treatment were confirmed.

The induction of systemic resistance in crops by exogenous application of SA represents a potentially valuable method in pathogen management strategies complementary to conventional control methods. Naturally induced SAR was not predictable in timing and level of expression and therefore, it could not be useful for agricultural practice. Hence, a novel approach of using synthetic signal molecules to induce SAR in crop plants had emerged. It is evident that SA is an important endogenous signal molecule involved in the transduction pathway and is required for the establishment of SAR (Shulaev *et al.,* 1995). SA also affects the lipid peroxidation, which plays a key role in initiating defense response (Anderson *et al.,* 1998) and induction of SAR in plants when challenged with pathogens (Maldonado *et al.,* 2002; Nandi *et al.,* 2004; Shah, 2005). The beneficial influence of SA on the growth and yield was also established by earlier workers (Meena *et al.,* 2001; Venkata Ratnam *et al.,* 2004).

SA has been shown to be a signalling molecule involved in both local defense reactions at infection sites and the induction of systemic resistance (Durner *et al*., 1997). The present study confirmed the systemic nature of resistance against the pathogen which is induced through exogenous application of SA. A similar result was made on the effect of SA on induction of systemic resistance by

Persaud *et al.* (2010) who mentioned that salicylic acid enhanced the resistance in rice plants against sheath blight disease and also increased the grain filling percentage.

In the present study, the cumulative effect of soil application (ZSS), chemical spray SA_1 and PS_2 spray not only minimized the disease incidence but also had positive influence on the biometrics and yield of ADT 36. This might be due to the positive interaction and synergism between $ZnSO_4$, Salicylic acid and potassium silicate in minimizing the disease incidence and enhancing the plant growth and grain yield of ADT 36.

Besides the adverse effect on the disease, the treatments with ZSS, SA_1 and PS2 had profound effect on the biochemical and enzymatic constituents of the plants and induced positive changes in infected plants under pot culture conditions. Disease reduction was due to the increased resistance to the disease by increased levels of inorganic compounds (Sugimoto *et al.,* 2009). It was therefore hypothesized that the increased conc. of macro - micro nutrient in plants due to the application of zinc sulphate and potassium silicate might have also induced an improvement of the physical properties of cells in rice plants.

Thus, the results of the present study have confirmed the potential possibility of using ZSS, SA_1 and PS_2 as alternative for chemical fungicides for the management of brown spot caused by *H. oryzae* and enhancing the crop yield without any deleterious impact on the environment and soil biota.

Table 1. Efficacy of ZS, SA, PS and *H.oryzae* inoculation on the disease incidence and biometrics of rice var. ADT 36 (Pot culture experiment)

T. No	Treatments	Disease incidence (%)	Per cent increase over control	Plant height (cm)	No. of tillers/ clump	No. of productive tillers/ clump	Panicle length (cm)
1	ZSS + ZSF_1 + ZSF_2	12.86	78.80	84.36	12.48	11.47	16.69
2	ZSS + SA_1 + SA_2	11.98	80.25	86.40	13.16	12.57	17.23
3	ZSS + PS_1 + PS_2	12.70	79.06	84.90	12.57	11.98	16.81
4	ZSS + ZSF_1 + SA_2	11.76	80.61	88.27	13.52	12.93	17.58
5	ZSS + SA_1 + ZSF_2	11.30	81.37	88.53	13.60	13.00	17.67
6	ZSS + SA_1 + PS_2	10.51	82.67	89.56	14.02	13.16	17.99
7	ZSS + PS_1 + SA_2	10.82	82.16	88.86	13.83	13.05	17.90
8	ZSS + PS_1 + ZSF_2	12.54	79.33	85.70	12.90	12.27	17.02
9	ZSS + ZSF_1 + PS_2	12.30	79.72	86.05	13.34	12.80	17.41
10	Carbendazim	15.47	76.14	82.75	12.06	11.28	16.84
11	Control	60.67	----	72.65	10.64	10.12	16.00
	C.D. (p=0.05)	1.19	---	0.12	0.10	0.05	0.04

Table 2. Efficacy of ZS, SA, PS and *H.oryzae* inoculation on yield parameters of rice var. ADT 36 (Pot culture experiment)

T. No.	Treatments	Thousand grain wt. (g)	Filled grain (%)	Grain yield (g/pot)	Straw yield (g/pot)
1	ZSS + ZSF_1 + ZSF_2	20.54	80.14	34.90	81.74
2	ZSS + SA_1 + SA_2	20.57	81.69	35.28	86.40
3	ZSS + PS_1 + PS_2	20.54	80.66	35.03	82.80
4	ZSS + ZSF_1 + SA_2	20.60	82.02	35.36	87.07
5	ZSS + SA_1 + ZSF_2	20.60	82.28	35.44	87.34
6	ZSS + SA_1 + PS_2	20.65	83.06	35.60	88.27
7	ZSS + PS_1 + SA_2	20.63	82.65	35.52	87.90
8	ZSS + PS_1 + ZSF_2	20.55	81.07	35.14	84.66
9	ZSS + ZSF_1 + PS_2	20.55	81.38	35.22	85.21
10	Carbendazim	20.54	74.80	30.86	79.42
11	Control	20.50	63.18	26.44	67.20
	C.D. (p=0.05)	0.11	0.13	1.32	1.01

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