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Response of Some Commercial Cultivars and Advanced Lines of Wheat against Karnal Bunt of Wheat and Its Management through Chemicals

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Abstract Wheat is the staple food crop for the people of Pakistan. Considerable losses in terms of crop yield are inflicted by Karnal bunt disease caused by *Tilletia indica*. The Research was under taken in the research area of Department of Plant Pathology, University of Agriculture, Faisalabad. There were 89 wheat advanced lines and commercials varieties which were screened against quarantine pathogen *Tilletia indica*. Five heads of each advanced lines and commercials varieties were manually inoculated by Hypodermal syringe. Primary and secondary sporadial suspension was prepared to inoculate the plants. The screening of 89 advanced lines of wheat revealed that and No lines were Highly resistant (Immune), 5 were Resistant, 21 Moderately resistant, 24 Moderately susceptible, 16 were susceptible and 6 lines and commercial Wheat varieties were Highly susceptible, respectively. In the second experiment for evaluation of chemicals for disease control, it was observed that Crest at their 40µg/ml and 60µg/ml dosage rates were the most effective fungicide in inhibiting the colony growth of *Tilletia indica* produced 2.30 mm and 2.13 mm diameter and Crest at 80µg/ml produce 1.92. Dolomite at 40ug/ml produces 1.71 mm diameter of inhibition zones of the fungus and Dolomite at 60ug/ml produces 1.30 mm diameter. Shelter however at 40 µg/ml dosage rate displayed the statistically effectiveness results like 1.93 mm. Thiomil proved to be less effective against the pathogen. Crest proved to be a best fungicide for disease control under in vitro conditions.

Keywords Karnal bunt disease, Tilletia indica Mitra (Mundkur) *Triticum aestivum* L., Fungicidal control

1. Introduction

Wheat (Triticum aestivum L.) is the main food crop and major source of nutrition for the people of Pakistan. It is used as a staple food in Pakistan and other countries. It plays a remarkable role in meeting the food requirements and economic stability of the country. In the GDP its share is about 3.1 %. According to Economic Survey of Pakistan, 2009-10 wheat was cultivated on an area of 9.042 million hectares. Production target of wheat was set 25 million tons in 2009-10, but the size of wheat crop was 23.864 million tons[5].

Karnal bunt was first detected in 1931 at Karnal in Haryana, India and hence it is called Karnal bunt[12]. It bears many names such as Karnal bunt, new bunt, partial bunt, incomplete bunt, Indian bunt and stinking smut.

Singh *et al*.,[16] reported that Karnal bunt was a disease of wheat, durum, rye and triticale (a hybrid of wheat and rye). Though the disease is native to South Asia but subsequently it has been reported from Iran, Syria, Afghanistan, Iraq, Mexico[8].

The pathogen infects the ovaries in the emerging wheat heads and converts the grains partially or completely into dark colored powdery masses of teliospores. The diseased fields emit a foul smell like that of rotten fish due to production of Trimethyl amine. Karnal bunt can reduce wheat yields. There is no estimate of losses due to this disease occurring in Pakistan; however survey in India conducted that years of heavy disease revealed a total loss of 0.5 percent, but in some fields where 89 percent of the kernels were infected, the yield losses ranged from 20-40 percent in highly susceptible varieties[6].

As the pathogen is soil seed and air-borne, it can penetrate locally into host plant, so application of spray fungicides is very critical. Epidemiological factors have great influence on the epidemic development of karnal bunt disease. Wheat is vulnerable to Karnal bunt fungus only during a 2-3 week windows of its' physiological development stages if the environmental conditions happen to be conducive during this short period for successful infection and the weather favorable for the disease

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development does not exist every year[18].

Karnal bunt (KB) of wheat, caused by Tilletia indica (Mitra) Mundkur, is responsible for minor yield losses and a fishy odour caused by the production of trimethylamine[11].

2. Materials and Methods

1. Screening of advanced lines/varieties received from Wheat Research Institutes, Faisalabad by artificial inoculation.

2. *In vitro* evaluation of fungitoxicants for optimizing the management of Karnal bunt disease of Wheat.

2.1. Screening of Germplasm/ Varieties against *Tilletia indica*

Teliospores from one year old bunted seeds was incubated on plain agar at 15-20℃ (Mishra et al., 2002). After 10-12 days germinating teliospores was shifted in a flask containing PDA in slanting position to produce the primary and secondary sporidia in mass culture[13]. Advanced germplasm lines and commercial wheat varieties sown in a single row of 3m length and all the plots were screened against the disease by incoulatin 5 heads of each variety. Test entries after boot inoculation with primary and secondary Sporidial suspension were tagged. Inoculated heads was hand threshed to calculate the bunted and healthy seeds. The level of resistance/susceptibility of each test entry were determined by a modified disease rating scale of Aujla *et al.*,[2]. The levels of resistance/susceptibility of the test cultivars were assessed by using the following modified disease rating scale of Aujla *et al.*,[2]. The incidence of the disease for each entry was calculated by using the following formula. The data of inoculated heads were recorded on percent grain infections.

$$\text{Disease incidence} = \frac{\text{No. of bunted grains in 10 spikelets}}{\text{Total number of grains in 10 spikelets}} * 100$$

Figure 1. Inoculated Wheat Field

Table 1. Rating scale used to determine level of resistance/ susceptibility

Disease rating scale	% Grain infection	Level of resistance or susceptibility
0	No infection on (panical)	Highly resistant
1	1 % or less grains bunted	Resistant
3	1.1-2 % of grains bunted	Moderately resistant
5	2.1-5 % of grains bunted	Moderately susceptible
7	5.1-10 % of grains bunted	Susceptible
9	More than 10 % of the grains bunted	Highly susceptible

2.2. *In vitro* Evaluation of Chemicals

The efficacy of four fungicides in inhibiting the colony growth of *Tilletia indica* was tested through inhibition zone techniques at 40 ppm, 60 ppm and 80 ppm[10]. Sporidial suspension was spread on the surface of the Petri plates containing PDA. With the help of cork borer a well was made in the center of each Petri plates to put the fungicide solution in each replicated Petri plates[10]. After incubation at 15℃ for 6 days, zone of the inhibition of mycelial growth made around the vicinity of the fungicidal suspension was recorded. The data were analysed statistically by subjecting it to analysis of variance and Least Significance Difference Test (LSD) Steel and Torrie,[14] to visualize the difference between the effects of various fungicidal spray treatments.

3. Results and Discussions

There were 89 advanced lines and Commercial varieties which were sown in single Row plot in experimental area. AS-2002 was the spreader in the field because in past disease were coming on this variety very easily. Regarding the approaches towards breeding for disease resistance, the screening of 89 advanced lines of wheat revealed that and No lines were Highly Resistant (Immune), 5 lines and commercial Wheat Verities were Resistant, 21 lines and commercial Wheat Varieties were Moderately Resistant, 24 lines and commercial Wheat Varieties were Moderately Susceptible, 16 lines and commercial Wheat Varieties were susceptible and 6 lines and commercial Wheat Varieties were Highly Susceptible, respectively.

Table 2. Level of resistance/susceptibility of advanced wheat lines and commercial cultivars against Karnal bunt disease of wheat

Grade in the disease rating scale and percent grain infection	Response of test line or cultivar against the disease	Advanced wheat lines	Commercial wheat cultivars
0 = No Infection at All	Highly Resistant Immune	No. advance Lines and Commercial Varities were Highally Resistat	-
1 = Less than 1% grain bunted	Resistant	V-08200, V-08212, V-087094, V-010296	PB-70
3 = 1.1-2% grain bunted	Moderately resistant	V-08064, V-08305, V-08308, V-08243, V-05082,V-04178, V-08158, V-05066, V-04022	Koh-97, Parwaz-94,Uqab-2000, Ufaq, Lassani-08, Shafaq-06, Bhakkar-02, Tecora-70, Borelag-95, Punjab-81, Punjab-85,Faisalabad-85
5 = 2.1-5% grain bunted	Moderately susceptible	V-088132, V-08211, V-08310, V-08335, V-09221, V-08008, V-09407, V-0BT005, V-06018, V-09272, V-09476, V-06111	MH-97, WL-711, Shalimar-88, Faisalabad-08, Inqalab-91, LYP-73, Manthar, CHK-86, Blue Silver. Farred
7 = 5.1-10% bunted grain	Susceptible	V-08081,TW076004, V-070067, V-0BT016	AS-2002, NR-388, PBW-343, PB-96, Rohtas-90, Iqbal-2000, Shehar-06, PAK-81, BWP-2000, CHK-97, Niab Wl-1
9 = More than 10 percent grain bunted	Highly susceptible	V-02192, V-085205	NB-378, Pasban-90, GA-2002, Kohsar-95

Table 3. Mean inhibition zones of colony of *Tilletia indica* by various fungicides at 3 dosages rates amended in PDA medium

Fungicides	Mean Inhibition zone (mm) at 3 dosage rates		
	40 ppm	60 ppm	80 ppm
Crest	2.30 h*	2.13 e	1.92 e
Dolomite	1.71 f	1.30 c	1.65 a
Shelter	1.93 ef	1.80 d	1.74 a
Thiomil	1.88 l	1.81 l	1.50 l
Water (control)	0.00	0.00	0.00

P=0.05 * Values having the same letters do not differ significantly at 5% level of Significance as determined by LSD = 0.2240 and EMS = 0.10

Table 4. ANOVA of Mean inhibition Zone *Tilletia indica* by Fungicides

SOV	D.F	S.S	M.S	F value	Prob>F
Treatment	3	1.487	0.496	9.2969^{S*}	0.003
Replication	2	0.485	0.242	4.5469	0.0212
Treatment x Replication	6	0.335	0.056	1.0469	0.4226
Error	24	1.280	0.05		0.053
Total	35	3.587			

Coefficient of Variation: 12.89 % P=0.05

In 2nd experiment for Evaluation of Chemicals against the Pathogen *Tilletia indica*. Dolomite and Shelter at all dosage rates were less effective than Crest and Shelter as a fungicide in laboratory produced 2.30, 2.13, 1.92 mm diameter inhibition zones respectively at 40 ug/ml, 60ug/ml and 80 ug/ml under invitro conditions. However, there was statistically difference between the effectiveness of Dolomite and Shelter at all dosage rates. Shelter at 40 µg/ml dosage rates and Crest at 80 ug/ml produced statistically same results like 1.93 mm and 1.92 mm in diameter inhibition zone of fungus. Thiomil fungicide produced statistically good results at 40ug/ml and 60 ug/ml dosage but there was less and not good result of Thiomil fungicide. Crest produced most economical results at all dosage rates by inhibition zone techniques. Control of all the replications was zero because there was not applied any fungicide in Petri plates and Fungus growth was maximum and no movement of zone towards the fungus. Karnal bunt disease of wheat has assumed on alarming situation in the Punjab during the previous two to three decades[4] and has been reported to cause, depending upon the cultivar affected, up to 30 percent grain losses[3,7]. This calls for control of disease either by the use of host resistance or through the use of chemotherapy. Since resistance to karnal bunt disease in available commercial wheat cultivars is absent[4], the chemotherapeutic control of the disease can be achieved by foliar application of fungicides at proper plant stage growth [7,15,16];. Foliar spray of fungicides may protect the plants from infections or eradicate the established infection[17]. The studies of this paper revealed that Dolomite and Shelter fungicides were not only the most and equally effective fungicides in inhibiting the in vitro colony growth of T. indica but these fungicides were also the most and equally effective for in vivo control of the karnal bunt infection of wheat grains. However, the protective applications of these fungicides were comparatively more effective in controlling wheat grain infection than their eradicative applications. Under the present situation of scarcity of resistance to Karnal bunt disease the control of the disease through chemotherapy is not uncommon[15] reported that Bavistine (Derosal-60) and Bayleton, when sprayed at boot leaf growth stage prior to inoculation (protective spray) controlled the karnal bunt disease. Singh and prasad[15] found that a single spray of Benomyl or Bevistine or Dithane M-45 at boot leaf growth stage was effective against karnal bunt of wheat.

4. Conclusions

Karnal Bunt of Wheat caused by Tilletia indica is becoming an alarming situation in whole area of Pakistan and this pathogen is destroying many commercial as well as advanced lines of Wheat and can cause Food Security Problems in Pakistan. AS 2002 was a spreader and a Susceptible Variety against Tilletia indica . When Screening was done to get source of resistance against Karnal Bunt then no variety/advanced line was resistant against this Pathogen. Weather conditions and other environmental factors helped the pathogen to cause the disease in wheat verities. Our mostly wheat varieties and advanced lines lack the gene of resistance for this pathogen. When process of chemical control was done then some fungicide concentration proved better results against this pathogen under in vitro conditions by Inhibition zone Technique. Some fungicides were less effective against this pathogen.

5. Recommendations

It is recommended that timely sowing and balanced use of Fertilizers leads to less infection of Karnal bunt of wheat. AS2002 is a spreader and a susceptible variety of wheat and it should not be sown. Proper quarantines measures should be taken for importing of wheat seed. Its chemical control in field is less effective and resistant varieties should be established. Proper Screening should be done before the release of new variety.

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