



EVALUATION OF WHEAT (*TRITICUM AESTIVUM* L.) VARIETIES/LINES AGAINST LOOSE SMUT (*USTILAGO TRITICI*) AND ITS MANAGEMENT BY SEED DRESSING FUNGICIDES

Syed Kamil Husnain, Sabir Hussain Khan, Shaukat Ali*, Muhammad Akhtar** and Muhammad Idrees*

ABSTRACT

Present study was conducted at Plant Pathology Research Institute, AARI, Faisalabad, Pakistan during 2012-14 to evaluate 92 wheat varieties/lines against loose smut caused by *Ustilago tritici* through artificial inoculation. Following augmented design RCBD each cultivar/line was planted in a single row of 3 meter length, with plant to plant and row to row distances of 5-10 and 15 cm, respectively. Germplasm was divided into six sets including four sets of 15 varieties/lines and two sets of 16 varieties excluding check varieties (WL-711, Morocco and Pak-81) which were sown in each set. Twenty spikes of each variety/line were inoculated with the spore suspension of loose smut using syringe method. V-ENO-310 exhibited high resistance showing 4.5 percent smut infected spikes, followed by Pasban-90 and 09b9110 with 9.3 and 7.8 percent smut infected spikes, respectively. Fifty varieties/lines were found moderately resistant having 10.8 to 20 percent smut infected spikes. On the other hand 26 varieties were found moderately susceptible having 20.23 to 30 percent smut infected heads, eight varieties/lines were found susceptible showing 30.7 to 48.1 percent smut infected heads and three check varieties (Morocco, WL-711 and Pak-81) were found highly susceptible showing more than 50 percent smut infected spikes. All the six fungicides (Dividend Star, Raxil Ultra, Score, Crest, Topsin-M and Hombre) were found equally effective as seed treatment for disease control.

KEYWORDS: *Triticum aestivum*; wheat; germplasm; wheat; screening; loose smut; *Ustilago tritici*; Pakistan.

*Plant Pathology Research Institute, AARI, Faisalabad.
**University of Agriculture, Faisalabad, Pakistan.

Article received on:

20/09/2014

Accepted for publication:

19/06/2015

INTRODUCTION

Wheat (*Triticum aestivum* L.) enjoys status of staple food among the cereal crops globally. Pakistan was ranked 8th in the world with average yield of 2787.4 kg per hectare and 26.45 million tons annual production from area of 8.69 million hectares (3). Wheat flour being compulsory part of daily diet is very important and cheap source of carbohydrates, proteins, vitamins and minerals.

Wheat crop is attacked by a large number of diseases among which rusts and smut are highly destructive and cause enormous damage. Loose smut caused by *Ustilago tritici* is prevalent throughout the world (15). Contaminated seed is the only source of perpetuation and loose smut causes yield losses upto 5-7 percent where farmers recycle their own seed (3, 12). Based on surveys conducted in northwestern India, 2-4

percent yield reduction of wheat from loose smut is estimated every year (9, 16). Presence of loose smut infection cannot be predicted until plant impregnated with the inoculum produces a spike characteristic symptoms i.e. early emergence and blackening of emerging spike. In a diseased plant spikelet and kernels of each spike there is smut infected mass instead of grains enveloped in greyish membrane which later on ruptures and black powdery mass i.e. teliospores emerge out (1). When these teliospores land on healthy flowers germinate and establish in pericarp and tissue of embryo before kernel matures. The mycelium then becomes inactive and remains dormant until the infected kernel germinates next year.

Environmental factors play a significant role in the development and spread of this disease

asteliospores germination occurs at 22 to 27 °C, where relative humidity ranges from 60-90 percent (4). Cold humid weather accompanied by heavy dew favors the development of disease at flowering stage. Generally it is believed that loose smut can cause 1 percent damage, but in northern parts of the country, damage may increase upto 20 percent depending upon wheat cultivars under cultivation (18). High severity of the disease during 1981-82, which caused damage upto 15 percent attracted the attention of researchers to find a way out, which may be use of chemical or cultivation of resistant variety. Significant control is observed by seed treatment with seed dressing fungicides and hot water seed treatment at 53°C for 15 minutes is also helpful in disease control. The use of tolerant varieties is the most suitable and economical method to control the disease.

The present study was planned to detect the resistant cultivars against loose smut caused by *Ustilago tritici* through artificial inoculation.

MATERIALS AND METHODS

An experiment was conducted in research area of Plant Pathology Section, Ayub Agricultural Research Institute, Faisalabad, Pakistan during 2012-14. Ninety two wheat varieties/lines were used from local sources (Wheat Research Institute Faisalabad, Regional Agricultural Research Institute, Bahawalpur, Arid Zone Research Institute, Bhakkar and Barani Agricultural Research Institute, Chakwal). Each cultivar/line

was planted in a single row of 3 meter length, with plant to plant and row to row distance of 5-10 cm and 15 cm, respectively following augmented design. Germplasm was divided into six sets including four sets of 15 varieties/lines and two sets of 16 varieties. Three check varieties (WL-711, Morocco and Pak-81) were sown in each set. The inoculum was prepared by dissolving one gram of teliospores in one liter distilled water (11). Syringe inoculation method was adopted by using 5 ml syringe with 24 hypodermic gauge to inoculate the spikes with fungus spores at leaf boot stage. Twenty spikes of each variety /line were inoculated with the spore suspension of loose smut. Inoculated spikes were tagged separately and were harvested during 2nd week of May and kept at room temperature and these were separately threshed in the month of June. These inoculated seeds were kept in paper bags and were stored at room temperature for next sowing season and were sown in the month of November in the same research area. Each cultivar/line was planted in a single row. Entries were sown in augmented design. When all the cultivars completed spiking, the diseased and healthy spikes were counted and disease incidence (percent spike infection) was calculated. Smut incidence and infected spikes were counted on basis of visual symptoms and the disease incidence was measured by using following disease rating scale against loose smut of wheat as developed by Ilyas *et al* (8).

Disease intensity

No symptoms on any plant
 0.1- 5% plants with smut infected spikes
 6-10% plants with smut infected spikes
 11-20% plants with smut infected spikes
 21-30% plants with smut infected spikes
 31-50% plants with smut infected spikes
 51-100% plants with smut infected spikes

Reaction

= Immune
 = Highly resistant (HR)
 = Resistant(R)
 = Moderately resistant (MR)
 = Moderately susceptible (MS)
 = Susceptible (S)
 = Highly susceptible (HS)

During the crop season 2012-13, susceptible wheat variety (WL-711) was sown in plot 3 x 2 meters in research area of Plant Pathology Research Institute, AARI, Faisalabad, Pakistan. When plants reached at anthesis stage, 300

spikes were inoculated by syringe method (11). The inoculated seeds of this variety were collected and stored for next year sowing. To find out the most effective fungicide as seed treatment against the loose smut recommended doses of six

fungicides including Davidend Star, Raxial Ultra, Topsin-M, Score, Hombre and Crest were used. Requisite quantity of each fungicide was dissolved in measured volume of water. Inoculated seeds of susceptible variety WL-711 were soaked in the solutions of each fungicide for recommended time period. In the month of November 2013-14 these inoculated and treated seeds were sown in the same area by keeping one treatment as a control, following RCBD. At the time of spike emergence, disease incidence percentage was recorded on the basis of smut infected spikes.

RESULTS AND DISCUSSION

Germplasm screening

The data (Table 1) revealed that no variety / line was found immune towards disease; one advance line (V-ENO-310) exhibited high resistance with 0.1-5 percent smut infected spikes, while two varieties/ lines (Pasban-90 and 09b9110) were found to be resistant with 6-10 percent smut infected spikes. On the other hand 55 varieties/lines (Fareed 2006, KOH-97, 9091, V-11154, V-8203, 10-BT-043, 10309, 8082, NN-400, V-2192/EN0310, V-10031, V-10306, NN-Gundm-1, Millat 2011, 8212, 9039, V-10110, 8068, V-11164, TW-961010, V-11153, V-9452, AS-2002, BHK-02, V-9082, Aqab-2000, V-11183, V-10104, V-7096, V-08314, V-11166, V-9087, V-10193, DH-31, Inqalab91, V-11149, TW-86014/

Iqbal, V-11156, TW-96011, B.W.P-97, Lasani 2008, V-10355, V-09136, Bullog-95, NR-399, Parwaz-94, V-10217, V-2003, V-10378, V-11178, LOB-9172, 7096, Chanab-2000, 10B9346 and V-11168) were rated as moderately resistant with 21-30 percent disease incidence. Other 26 varieties/lines (Punjanad 2001, Punjab2011, ARRI2011, ASS-11, Inqalab-2000, 8118, 9172, Shalimar-88, Seher 2006, 09-BT-043, NE, 10296, TW-96009, NN-Gundm-2, V-7894, V-10297, 6C002, TW-96010, 8171, 10C033, 10317, HD.29, 9006, Inqalab-91, V-11160 and 9137) behaved as moderately susceptible, whereas eight varieties/ lines (2003, Fareed-2006, 9C037, Miraj 2008, Shafaq 2006, Faisalabad 2008, 9110 and 9346) were found to be susceptible and only three check varieties/lines remained highly susceptible against the disease (Table 2).

Determination of resistant sources against loose smut disease is a common practice which reveals that sources of resistance are not Spartan in wheat germplasm. Beniwal *et al.* (4) reported that WH283, WH 291, WH 542, Raj. 3765, PBW 343 and PBW 373 and a durum variety WH 896 exhibited multiple disease resistance. Zia Ullah *et al.* (18) reported that Inqalab-91, CRS-1, Punjab-96, Faisal-85, WP-95, Derawar-97, and MH-97 had a susceptible reaction, whereas Iqbal-2000, Uqab-2000, and Bwp-2000 were resistant. Some 28 entries were free of loose smut infection. Rashmi *et al.* (13).

Table 1. Disease incidence in wheat germplasm against loose smut (*Ustilago tritici*).

Sr. No.	Varieties/lines		Disease incidence (%)	Difference among varieties/lines	Disease reaction
1.	Maroco	V	54.03	A	HS
2.	WL-711	V	53.63	Ab	HS
3.	Pak-81	V	51.23	Ab	HS
4.	2003	L	48.10	B	S
5.	Best Fareed	V	37.50	C	S
6.	9C037	L	34.50	Cd	S
7.	Miraj 2008	V	34.10	Cd	S
8.	Shafaq 2006	V	32.70	Cd	S
9.	Faisalabad-2008	V	31.70	Cde	S
10.	9110	L	30.90	Cdef	S
11.	9346	L	30.7	Cdef	S
12.	Punjanad 2001	V	30.00	Cdefg	MS
13.	Punjab2011	V	28.1	Defgh	MS

14.	ARRI2011	V	28.00	Defgh	MS
15.	ASS-11	V	27.10	Defghi	MS
16.	Inqalab-2000	V	27.00	Defghi	MS
17.	8118	L	26.63	Defghij	MS
18.	9172	L	25.40	Efghijk	MS
19.	Shalimar-88	V	24.76	Efghijkl	MS
20.	Seher 2006	V	24.63	efghijklm	MS
21.	09-BT-043	L	24.60	efghijklmn	MS
22.	NE	L	24.30	efghijklmno	MS
23.	10296	L	23.73	fghijklmnop	MS
24.	TW-96009	L	23.47	fghijklmnopq	MS
24.	NN-Gundm-2	V	23.17	fghijklmnopq	MS
26.	V-7894	L	23.07	fghijklmnopq	MS
27.	V-10297	L	22.60	ghijklmnopqr	MS
28.	6C002	L	22.10	hijklmnopqrs	MS
29.	TW-96010	L	21.77	hijklmnopqrst	MS
30.	8171	L	21.23	hijklmnopqrstu	MS
31.	10C033	L	21.17	hijklmnopqrstuv	MS
32.	10317	L	21.03	hijklmnopqrstuvw	MS
33.	HD.29	L	21.00	hijklmnopqrstuvwx	MS
34.	9006	L	20.83	hijklmnopqrstuvwxy	MS
35.	Inqalab-91	V	20.80	hijklmnopqrstuvwxy	MS
36.	V-11160	L	20.70	hijklmnopqrstuvwxyz	MS
37.	9137	L	20.23	hijklmnopqrstuvwxyzA	MS
38.	Fareed 2006	V	20.00	ijklmnopqrstuvwxyzA	MR
39.	KOH-97	V	19.80	ijklmnopqrstuvwxyzAB	MR
40.	9091	L	19.43	ijklmnopqrstuvwxyzABC	MR
41.	V-11154	L	19.20	ijklmnopqrstuvwxyzABC	MR
42.	V-8203	L	19.00	jklmnopqrstuvwxyzABCD	MR
43.	10-BT-043	L	18.87	klmnopqrstuvwxyzABCDE	MR
44.	10309	L	18.83	klmnopqrstuvwxyzABCDE	MR
45.	8082	L	18.83	klmnopqrstuvwxyzABCDE	MR
46.	NN-400	L	18.77	klmnopqrstuvwxyzABCDE	MR
47.	V-2192/EN0310	L	18.57	klmnopqrstuvwxyzABCDEF	MR
48.	V-10031	L	18.40	klmnopqrstuvwxyzABCDEF	MR
79.	V-10306	L	18.40	klmnopqrstuvwxyzABCDEF	MR
50.	NN-Gundm-1	V	18.37	klmnopqrstuvwxyzABCDEF	MR
51.	Millat 2011	V	18.30	lmnopqrstuvwxyzABCDEF	MR
52.	8212	L	18.13	lmnopqrstuvwxyzABCDEF	MR
53.	9039	L	18.03	lmnopqrstuvwxyzABCDEF	MR
54.	V-10110	L	17.90	lmnopqrstuvwxyzABCDEF	MR
55.	8068	L	17.83	lmnopqrstuvwxyzABCDEF	MR
56.	V-11164	L	17.60	lmnopqrstuvwxyzABCDEF	MR
57.	TW-961010	L	17.37	mopqrstuvwxyzABCDEF	MR
58.	V-11153	L	16.80	mopqrstuvwxyzABCDEF	MR
59.	V-9452	L	16.67	nopqrstuvwxyzABCDEF	MR
60.	AS-2002	V	16.30	pqrstuvwxyzABCDEF	MR
61.	BHK-02	V	16.10	pqrstuvwxyzABCDEF	MR
62.	V-9082	L	16.10	pqrstuvwxyzABCDEF	MR
63.	Aqab-2000	V	15.90	pqrstuvwxyzABCDEF	MR
64.	V-11183	L	15.70	qrstuvwxyzABCDEF	MR
65.	V-10104	L	14.90	rstuvwxyzABCDEF	MR
66.	V-7096	L	14.80	stuvwxyzABCDEF	MR
67.	V-08314	L	14.50	stuvwxyzABCDEF	MR
68.	V-11166	L	14.30	stuvwxyzABCDEF	MR
69.	V-9087	L	14.30	stuvwxyzABCDEF	MR

70.	V-10193	L	14.10	tuvwyzABCDEFGH	MR
71.	DH-31	L	14.07	uvwxyzABCDEFGH	MR
72.	Inqalab 91	V	14.03	vxyzABCDEFGH	MR
73.	V-11149	L	13.60	yzABCDEFGH	MR
74.	TW-86014/Iqbal	L	13.50	yABCDEFGH	MR
75.	V-11156	L	13.40	yABCDEFGH	MR
76.	TW-96011	L	13.06	yABCDEFGH	MR
77.	B.W.P-97	V	13.00	yABCDEFGH	MR
78.	Lasani 2008	V	13.00	yABCDEFGH	MR
79.	V-10355	L	12.90	yABCDEFGH	MR
80.	V-09136	L	12.90	yABCDEFGH	MR
81.	Bullog-95	V	12.80	zABCDEFGH	MR
82.	NR-399	L	12.56	ABCDEFGH	MR
83.	Parwaz-94	V	12.40	ACDEFGHI	MR
84.	V-10217	L	11.90	BCDEFGHI	MR
85.	V-2003	L	11.90	CEFGHI	MR
86.	V-10378	L	11.80	CEFGHI	MR
87.	V-11178	L	11.70	CEFGHI	MR
88.	LOB-9172	L	11.50	CEFGHI	MR
89.	7096	L	11.23	DEFGHI	MR
90.	Chenab-2000	V	11.20	EFGHI	MR
91.	10B9346	L	10.80	FGHI	MR
92.	V-11168	L	10.80	FGHI	MR
93.	Pasban-90	V	9.30	GHI	R
94.	09b9110	L	7.80	HI	R
95.	V-ENO-310	L	4.50	I	HR

* Mean sharing similar letters are statistically non-significant, Variety = V, Line = L, Highly susceptible = HS, Susceptible = S, Moderately resistant = MR, Resistant = MR, Highly resistant = HR

Table 2. Number of wheat varieties/lines showing disease incidence in wheat germplasm against loose smut (*Ustilago tritici*)

Disease incidence %	Reaction		No of varieties/lines	Varieties/lines
No symptoms on any plant	Immune	I	0	
0.1-5% plants with smut infected spikes	Highly resistant	HR	1	V-ENO-310
6-10% plants with smut infected spikes	Resistant	R	2	Pasban-90, 09b9110
11-20% plants with mottled spikes	Moderately resistant	MR	55	Fareed- 2006, KOH-97, 9091, V-11154, V-8203, 10-BT-043, 10309, 8082, NN-400, V-2192/EN0310, V-10031, V-10306, NN-Gundm-1, Millat-2011, 8212, 9039, V-10110, 8068, V-11164, TW-961010, V-11153, V-9452, AS-2002, BHK-02, V-9082, Aqab-2000, V-11183, V-10104, V-7096, V-08314, V-11166, V-9087, V-10193, DH-31, Inqalab-91, V-11149, TW-86014/Iqbal, V-11156, TW-96011, B.W.P-97, Lasani-2008, V-10355, V-09136, Bullog-95, NR-399, Parwaz-94, V-10217, V-2003, V-10378, V-11178, LOB-9172, 7096, Chanab-2000, 10B9346, V-11168
21-30% plants with smut infected spikes	Moderately susceptible	MS	26	Punjanad-2001, Punjab-2011, ARRI-2011, ASS-11, Inqalab-2000, 8118, 9172, Shalimar-88, Seher -2006, 09-BT-043, NE, 10296, TW-96009, NN-Gundm-2, V-7894, V-10297, 6C002, TW-96010, 8171, 10C033, 10317, HD.29, 9006, Inqalab-91, V-11160, 9137
31-50% plants with smut infected spikes	Susceptible	S	8	2003, Best Fareed, 9C037, Miraj-2008, Shafaq-2006, Fsd-2008, 9110, 9346
51-100% plants with smut infected spikes	Highly susceptible	HS	3	Maroco, WL-711, Pak-81
Total			95	

screened 539 wheat lines for resistance against loose smut using artificial inoculation and found some 28 entries free of loose smut infection, while Singh *et al.* (14) screened 931 advanced lines of wheat against loose smut and found that 99 lines were resistant to loose smut. If the resistance of above mentioned sources is incorporated into high yielding varieties/advanced lines can provide a tool for successful management of loose smut of wheat.

Table 3. Response of seed dressing fungicides against loose smut of wheat

No.	Fungicide	Active ingredient	Dose	Disease incidence	Percent Treatments/control
1	Davidend Star	Difenoconazole+ Cyproconazole	4.0ml/kg	0.71B	1.50
2	Raxial ultra 120 FS	Tebuconazole	1cc/kg seed	0.44B	0.93
3	Topsin-M 70 WP	Thiophenatemethyle	2g/kg seed	0.78B	1.65
4	Score 250 EC	Difenoconazole	1cc/kg seed	0.81B	1.71
5	Hombrae	Tebuconazole+ Imidacloprid	4.0ml/kg	0.87B	1.84
6	Crest	Carbendazim	2cc/kg	1.28B	2.71
7	Control			47.23A	

*Means sharing similar letters are statistically non-significant.

The finding of this study are somewhat similar to that of other studies conducted in other locations previously. Srivastava *et al.* (17) studied the efficacy of tebuconazole and reported that it gave excellent control of loose smut. Hussain *et al.* (7) reported that 5 systemic seed dressings tested against *Ustilago tritici* (*segetum* var.), Raxil-2 DS (tebuconazole) @ 2 g/kg seed gave 100 percent control in both test years (1988-89 and 1989-90). Difenoconazole @.15 percent is effective against loose smut of wheat (6). Loose smut is an important wheat disease. Due to its seed borne nature, it is easily amenable to systemic fungicides as dry seed treatment. However, due to high cost of chemical seed treatments and environmental hazards, breeding smut resistant varieties remains the ideal method to contain the disease.

Genetic material exhibiting resistance against the disease indicates the pathway to breeding programme for discovering resistant cultivars against the disease. So firstly lines exhibiting resistance should be incorporated in breeding programme for evolution of new genetic material and also fungicides used for the management of

Evaluation of seed dressing fungicides against loose smut of wheat

All the fungicides evaluated against loose smut of wheat as seed treatment were equally effective against the disease and statistically at par but significantly reduced the disease as compared to control (Table 3).

diseases should be recommended to the farmers for getting maximum grain yield.

REFERENCES

1. Agrios, G. N. 2005. Plant Pathology. Burlington: Elsevier Academic Press (5th Ed.) p. 584-588.
2. Anon. 2013. FAOSTAT data[<http://faostat.fao.org/>]
3. Anon.1992. Project Director's Report 1991-92. All India Coordinated Wheat Improv. Proj.Directorates of Wheat Res. Karnal. p.8.
4. Beniwal, M.S., K.S. Virk and R. Singh.1999. Relative resistance of commercially grown wheat varieties of north-western plain zone of India against important diseases. Crop Res. Hisar. 18: 296-298.
5. Druzhin, A. E and V.A. Krunov. 2000. Influence of environmental conditions of infection of spring wheat by loose smut in Volga region. Russian Agric. Sci.3:16-19.
6. Grewal, A. S., G. S. Nanda and K. Baljinder. 1996. Efficacy of Dividend, a new systemic fungicide in controlling loose smut of wheat. Plant Dis. Res.11:188-190

7. Hussain, S., A.H. Tariq and A. Saleem.1991. Chemical control of loose smut of wheat. Pak. J. Phytopath. 3: 72-73.
8. Ilyas M.B., I. A. Mirza and A. B.Munir.1990. Evaluation of inoculation methods and screening of wheat against loose smut. Pak. J. Agric. Sci.27(3):252.
9. Joshi, L.M., D.V. Singh, and K.D. Srivastava.1985. Monitoring of wheat disease during 1984-85. In: Proc. 24th All India Wheat Res. Workers' Workshop, GBPUAT, Purnnagar.
10. Khanzada, A. K. and M. Aslam.1982. Reaction of wheat varieties to loose smut. National Seminar on Wheat Research and Production. Islamabad, Pakistan.
11. Mishra, R. P.,S.P. Tiwa and M.N. Khare. 1990. Studies on loose smut of wheat. Testing of resistance and susceptibility for wheat varieties to *Ustilago tritici* (Pers) Rostr under artificial inoculation. Ind. J. Myco. Plant Pathol. 20:171-173.
12. Ramdani, A., M. Jlibene and M. Boulif. 2004. Survey of wheat diseases in the north west region of Morocco during 1997-98. Al Awamia. 111: 33-40
13. Rashmi, A., K. D. Srivastava, P. Bhadur, D.V. Singh and R. Aggarwal. 1998. Resistance to loose smut and rust in wheat. Rachis. 17: 63-64.
14. Singh, D.P., A.K. Sharma, M.S. Saharan, J. Kumar and J. Shoran. 2005. Status of resistance in popular wheat cultivars against major diseases in India. Integrated plant disease management Challenging problems in horticultural and forest pathology, Solan, India, 14 to 15 November. p. 279-287.
15. Sinha, A. P. 1997. Effect of loose smut infection on the growth and morphology of wheat plants. Agric. Sci. Digest Karnal. 17: 209-212 .
16. Srivastava, K. D., D.V. Singh, R. Aggarwal, P. Bahadar and S. Nagarajan. 1992. Occurrence of loose smut and its sources of resistance in wheat. Ind. Phytopath. 45:111-112.
17. Srivastava, K. D., D. V. Singh, R. Aggarwal, A.K. Dixit and P. Bahadur.1997. Bioefficacy and persistence of tebuconazole against loose smut in wheat. Ind. Phytopath. 50:434-436.
18. Zia Ullah, H.M., A.H. Tariq, S. Ahmad, M.A. Hussain and L.H. Akhtar. 2003. Prospectus for resistance to loose smut of wheat in southern Punjab. Ann. Wheat News. p:89-92.

CONTRIBUTION OF AUTHROS

Syed Kamil Husnain	Conducted the experiment, analysed the data and prepared the writeup
Sabir Hussain Khan	Supervised the experimental work
Shaukat Ali	Planned the research and reviewed the article
Akhtar Hameed	Helped in experimental work
Muhammad Idrees	Reviewed the literature