

## SELECTION OF BARLEY GERMPLASM RESISTANT TO SPOT BLOTCH

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### Abstract

Spot blotch is a common disease of barley and wheat caused by *Bipolaris sorokiniana* (*Cochliobolus sativus*). The disease is found where ever barley is grown. Total 130 lines of barley (provided by ICARDA – CIMMYT Wheat Improvement Program) were screened for resistance to spot blotch caused by *Bipolaris sorokiniana* under controlled and natural conditions at NARC (National Agricultural Research Centre) Islamabad, Pakistan. Out of 130 lines, 7 barley lines viz. Jet, Bey, Forrajera, Beecher, Fiaz, Manchuria and Munch Palidum showed single gene base resistance under controlled and field conditions. However four lines including ETHIOPIA (CI 08755), ETHIOPIA (CI 20019), Mac Key-4 and Mac Key-9 gave maximum disease reaction (MRMS) at seedling stage under controlled conditions while they gave low disease reaction (R) at adult stage. These four lines may have more than one gene involvement and can further be exploited as a source of resistance to spot blotch in the breeding programme.

### Introduction

Barley (*Hordeum vulgare*) is probably the most important cereal crop grown under severe environmental stress (Harlan, 1968). Foliar and seed borne diseases are prevalent in all barley growing areas. Spot blotch is one of the diseases of barley world-wide caused by *Bipolaris sorokiniana* (*Cochliobolus sativus*). It causes significant yield losses under warm and moist environments (Clark, 1979). Early disease infection often affects the stand establishment of the barley crop and hence greatly reduces tiller number and productivity. This situation is very common in areas where barley is cultivated annually (Yahyaoui, 2002). Barley leaf blight incidence in Maghreb countries had been reported in 1999 as seriously damaging barley crops all over the region (Sayoul *et al.*, 1999) and cited 30–80% yield losses. In upper Midwest of the United State, a yield loss of 10–30% is reported when weather conditions are conducive for the disease development (Mathre, 1997). Yield losses in Kazakhstan in epidemic periods can reach 25–45%, while in Russia it is reported as 41.4% (Hasanov, 1992).

The pathogen attacks all parts of the plant in the form of oval brown necrosis, with the typical presence of a black area in the centre of the spot. It occurs jointly with net blotch (*Pyrenophora teres*). The management having fungicide protection is very costly and difficult in its application; therefore the use of resistant cultivars is the preferred means to control this disease. Current barley cultivars do not have adequate levels of resistance to *Cochliobolus sativus*. Resistance of commercial cultivars may loose its effectiveness because of genetic changes in the pathogen population. So the resistance breeding through the selection of the appropriate parents is used as the main strategy to reduce yield losses. In order to identify new sources of resistance in International Barley Nurseries received from ICWIP have been tested by *In vitro* and *In vivo* methods under this study. That may be helpful in the breeding efforts where attempt to combine disease resistance with high productivity is required.

## Materials and Methods

**Lab screening:** The inoculum of single spore culture of the most aggressive isolate of *B. sorokiniana* isolated from barley was increased on PDA (potato dextrose agar medium). Test tubes measuring (20 cm x 3 cm) were prepared by filling 1/4<sup>th</sup> of cotton in the bottom of the tube. Distilled water (20 ml) was added in each tube and tubes were covered with aluminium foil and autoclaved. The barley seeds (three seeds/tube) were surface disinfected with 1% Clorox solution for one minute and placed on the moist cotton swab in the test tubes. With the help of cork borer one disc of 5 mm of fungal isolates was taken having  $3.2 \times 10^4$  conidia/ disc and was placed adjacent to the seeds. After inoculation tubes were again covered with aluminium foil and were placed in growth chamber at 25°C for incubation. The data was recorded upon the appearance of spots on the leaves by 0-5 scale, where 0 = no symptoms (immune), 1 = 1-5% spots on leaves (R), 2 = 6-20% spots on leaves (MR), 3 = 21 – 40% spots on leaves (MRMS), 4 = 41 – 60% (MS), 5 = more than 60% (S) (Ifikhar *et al.*, 2007).

**Field screening:** A total of 130 lines supplied by ICARDA–CIMMYT Wheat Improvement Program (ICWIP) were evaluated in the field of NARC, Islamabad under natural conditions for spot blotch resistance in the year 2006. The germplasm to be screened was planted with single row having 5m with a row to row distance of 60cm. The data was recorded at grain formation stage on 0-5 scale, where 0 = Immune, 1 = R (resistant), 2 = MR (moderately resistant), 3 = MRMS (moderate resistant moderate susceptible), 4 = MS (moderate susceptible) and 5 = S (susceptible).

## Results and Discussion

One hundred thirty lines of barley received from ICWIP, were evaluated against *Bipolaris sorokiniana* at NARC experimental field and CDRP, NARC lab during 2006. The results revealed that 7 barley lines viz. Jet, Bey, Forrajera, Beecher, Fiaz, Manchuria and Munch Palidum were resistant at seedling as well as at adult stage (Table 1). These resistant lines having the single gene resistance can serve as a good source of resistance in barley breeding programme but they can never be used as a cultivar for long term because on exposure of virulence in nature, it may cause a big epidemic with huge loss of production. Thirty lines showed moderate resistance, out of which 14 lines gave 2 (MR) reaction under both (control and field) conditions and 16 lines gave 2 (MR) reaction under *In vitro* conditions but in field they gave 1 (R) reaction on 0-5 scale, that could be due to high inoculum pressure accompanied with conditions favouring the disease development as observed in a study where the severity of the isolates exhibited aggressive reaction *In vitro* conditions as compared to field conditions (Asad *et al.*, 2007) and also supported by Jain & Prabhu (1976) that the success of the pathogen is a need of its interaction with the conditions and the inoculum pressure. Same was the case with 4 varieties viz., ETHIOPIA (CI 08755), ETHIOPIA (CI 20019), Mac Key-4 and Mac Key-9 which gave 3 (MRMS) reaction at seedling stage under controlled conditions while they gave 1 (R) reaction at adult stage under field conditions. In these four lines more than one gene for resistance may be involved as these lines giving maximum reaction *In vitro* conditions followed by low disease reaction in field conditions and these lines can be exploited further. The remaining lines fall in the category of partial susceptibility and susceptibility.

**Table 1. *In vivo* and *In vitro* screening of barley germplasm for resistance to Spot blotch (*Bipolaris sorokiniana*) during 2006-07.**

<b>S. No.</b>	<b>Genotypes</b>	<b><i>In vivo</i> score</b>	<b><i>In vitro</i> score</b>
1.	Armelle	3	4
2.	Astrix	3	4
3.	Athene	5	5
4.	Igri	1	2
5.	La mastia	2	3
6.	Osiris	1	2
7.	Pirate	4	5
8.	Digger	2	2
9.	Rithane.0.3	3	4
10.	Line9-26-F27	3	4
11.	Granado	1	2
12.	TREBI	1	2
13.	JET	1	1
14.	KITCHIN	3	4
15.	OSIRIS	2	3
16.	STEUDELLE	1	2
17.	BEY	1	1
18.	ATLAS46	2	2
19.	LA MESITA	1	2
20.	MODOC	2	2
21.	FORRAJERA	1	1
22.	Klages	2	3
23.	ABYSSININ	3	4
24.	W12291	2	2
25.	SLB22-82/H.spont.38-3	1	2
26.	Beecher	1	1
27.	Carvette	2	2
28.	C19214	2	4
29.	Prior	3	4
30.	Skiff	2	5
31.	Rihane.03	3	5
32.	MARTIN	3	3
33.	Beecher	3	3
34.	Pirka	2	3
35.	C12330-Marchuria	1	2
36.	Fiaz	1	1
37.	Cross96-55	1	2
38.	TIFANG	1	2
39.	MUNCHURIA	1	1
40.	MUNCH.PALLIDUM	1	1
41.	CI 04207	2	3
42.	MUNCH.PALLIDUM	3	4
43.	ETHIOPIA	3	3
44.	ETHIOPIA	2	4

Table 1. (Cont'd.).

S. No.	Genotypes	<i>In vivo</i> score	<i>In vitro</i> score
45.	ETHIOPIA	2	3
46.	ETHIOPIA	1	3
47.	ETHIOPIA	1	3
48.	ETH GAW 80-4	1	2
49.	RUSS94-1	5	5
50.	Litani	3	4
51.	Tichidrit	2	3
52.	Saida	3	4
53.	Tokak	2	4
54.	Rihane.03	2	3
55.	Tipper/ISO10R	3	4
56.	Mac Key-1	3	5
57.	Mac Key-2	2	3
58.	Mac Key-3	2	4
59.	Mac Key-4	1	3
60.	Mac Key-5	1	2
61.	Mac Key-6	2	3
62.	Mac Key-7	1	2
63.	Mac Key-8	2	2
64.	Mac Key-9	1	3
65.	Mac Key-10	2	2
66.	Mac Key-11	2	3
67.	Mac Key-12	3	4
68.	Mac Key-13	2	2
69.	Mac Key-14	2	2
70.	Mac Key-15	2	2
71.	Gloria'S'/Copal'S'	1	2
72.	SARAROOD-1	3	4
73.	CI 05401	2	3
74.	CI 06311	4	5
75.	CI 09820	2	3
76.	CI 00739	1	2
77.	CI 01243	3	3
78.	CI 04795	2	4
79.	CI 04502	2	4
80.	CI 04979	3	3
81.	PROCTOR	3	3
82.	CODE 65	3	4
83.	CI 09214	3	3
84.	TENN 61-119	2	2
85.	CI 09214	4	5
86.	A.ABIAD	3	3
87.	W.HASSA	3	3
88.	CI 02330	4	5
89.	INAT 103	4	5

Table 1. (Cont'd.).

S. No.	Genotypes	<i>In vivo</i> score	<i>In vitro</i> score
90.	INAT 104	4	5
91.	ESAK 17	2	3
92.	ESAK 33	2	3
93.	CI 04795	3	4
94.	CI 04922	3	3
95.	NORODAL	3	4
96.	GUNHILD	2	3
97.	CLERMONT	3	5
98.	RIHANE.03	3	4
99.	WI 2291	4	5
100.	TADMOR	4	5
101.	TIFANG	3	4
102.	CANADIAN LAHE SHORE	2	3
103.	ATLAS46	3	4
104.	ROJO	4	4
105.	COAST	4	5
106.	MANCHURIN	4	5
107.	MING	2	3
108.	CI 9819	3	4
109.	ALGERIAN	5	5
110.	KOMBAR	3	4
111.	CI11458	2	3
112.	CI 5791	2	3
113.	HARBIN	3	4
114.	CI 7584	2	2
115.	PRATO	2	2
116.	MANCHURIN	3	4
117.	CI 5822	2	3
118.	CI 4922	4	4
119.	HAZERA	4	5
120.	CAPE	4	5
121.	BEECHER	3	4
122.	RIKA	3	3
123.	HECTOR	2	4
124.	FR 926-77	2	5
125.	ER/Apm//Akrash	2	3
126.	Rotha/3/Anoidium/Arig8//Rt013	1	2
127.	Marar/4/CompCr229//As46/Pro/3/Srs	2	2
128.	Misratch/R023	3	4
129.	LB.KanMehterzai/K-273	3	3
130.	CARBO		

**0-5 Scale:** 0= Immune, 1= Resistant (R), 2= Moderate resistant (MR), 3= Moderate resistant Moderate susceptible (MRMS), 4= Moderate susceptible (MS) and 5= Susceptible (S)

The results indicate that the *In vitro* method can be adopted for preliminary screening of the varieties/ germplasm and is also considered to be more convenient and less time consuming before going to field as observed by Iftikhar *et al.*, (2007). This satisfactory coincidence of data obtained from the type reaction of seedling under controlled conditions and disease severity in adult plant stage can be helpful in providing the good source of resistance against spot blotch in breeding programme to have the disease resistance with combination of high productivity.

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