



## Genetics of stripe rust resistance in two barley (*Hordeum vulgare* L.) genotypes

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### Abstract

Stripe rust inflicted by the pathogen *Puccinia striiformis* Westend. f. sp. *hordei* Eriks can cause marked grain yield reduction and deterioration of the malting quality in barley. Two barley genotypes, DWRB137 (DWR28/DWRUB64) and DWRB143 (DWRB73/DWR83) were identified as resistant (R) for three stripe rust races viz., 57 (0S0), M (1S0) and Q (5S0) at seedling and adult plant stages. These genotypes were crossed with two stripe rust susceptible (S) barley cultivars, RD2035 and Lakhan to study mode of inheritance of stripe rust resistance. All the F<sub>1</sub> plants showed resistance against stripe rust race, 57 (0S0) indicating that the resistance (R) is dominant. The F<sub>2</sub> generations derived from the crosses namely, Lakhan/DWRB137, Lakhan/DWRB143, RD2035/DWRB137 and RD2035/DWRB143 were tested with inoculum of race, 57 (0S0) under artificially inoculated conditions. The observed frequency of segregants in each F<sub>2</sub> generation fit well in a theoretical ratio of 3(R):1(S) ( $\chi^2_{(T)} < 0.01$ ) indicating that the resistance against the tested race is controlled by a single dominant gene in the genotypes, DWRB137 and DWRB143. To validate the F<sub>2</sub> hypothesis, the F<sub>3</sub> progenies were also tested under above described conditions and followed discrete segregation of 1 (R): 2 (Segregating): 1(S) ratio in all the four crosses. Test of allelism was also conducted to establish the identity of resistance gene(s) present in the resistant genotypes. The F<sub>2</sub> population derived from DWRB137/DWRB143 (R × R) cross fit to 15(R):1(S) ratio showing that the two genotypes had different resistance genes.

**Key words:** Stripe rust, barley, inheritance, test of allelism

Stripe rust, caused by *Puccinia striiformis* is an economically important disease of wheat, barley, rye,

triticale and other graminaceous members (Wellings 2011). The pathogens that cause stripe rust of wheat (*Puccinia striiformis* f. sp. *tritici*) and barley (*P. striiformis* f. sp. *hordei*) are considered to be *formae speciales* (special forms) and even the host ranges are almost similar of these separate forms (Derevnina et al. 2015). Wheat and barley rust pathogens belong to the genus *Puccinia*, family Pucciniaceae, order Uredinales and class Basidiomycetes. Systematic evaluation of wheat varieties against different rust pathotypes and rust race analysis was initiated somewhere in 1930s in wheat (Tomar et al. 2014), while the structured stripe rust resistance work on barley gained attention in 1942 (Jain and Agrawal 1964; Murty 1942).

Barley (*Hordeum vulgare* L.) stripe rust (*Puccinia striiformis* Westend. f. sp. *hordei* Eriks.) is one of the most devastating fungal diseases and may cause marked grain yield reduction coupled with loss of quality due to the shrivelled grains. The grain yield losses have been reported from 30-66% under different studies in barley. The disease is a serious concern in northern hills, foothills and in the states of Haryana, Punjab, western Uttar Pradesh and Rajasthan, which occupy more than 50% acreage under barley crop primarily for malting and feed purposes. Host plant resistance is one of the effective, sustainable, eco-friendly and cost effective approach for rust management in cereal crops. However, for resistance breeding, the knowledge of available resistant sources, nature of inheritance

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and identity of resistance genes is essential. Though a number of studies have been conducted on stripe rust resistance in wheat, meager information is available on the genetics of stripe rust resistance in barley. Some previous studies were carried out during nineteen sixties (Bahl and Bakshi 1963; Bakshi et al. 1964; Luthra 1966) involving the then resistance sources like EB410, EB438 and other EB series in barley, which have now become obsolete and pathogen race dynamics has also changed. Therefore, the present investigation was carried out to study the inheritance and allelic relationship of stripe rust resistance in two presently effective resistance sources, DWRB137 and DWRB143 using the prevalent stripe rust pathotype 57 (0S0).

Two barley genotypes, DWRB137 (DWR28/DWRUB64) and DWRB143 (DWRB73/DWR83) were evaluated with the barley stripe rust races, 57 (0S0), M (1S0) and Q (5S0) at seedling and adult plant stages (Anonymous 2015). Both the genotypes showed high degree of resistance (R) at seedling (0;) and adult plant stages (0) in field under artificially inoculated conditions (Anonymous 2015). Therefore, crosses of each of these genotypes were made during *rabi*, 2015-16 with two susceptible barley cultivars *viz.*, RD2035 and Lakhan to study the mode of inheritance of stripe rust resistance. A cross between both the resistant genotypes, DWRB137 and DWRB143 (R × R) was also made for the test of allelism. Part of the generated F<sub>1</sub> seeds were multiplied during off-season nursery at ICAR-IIWBR, Regional Station, Dalang Maidan, Himachal Pradesh and the parents, F<sub>1</sub> and F<sub>2</sub> generations were grown in rows at 30 cm apart with plant to plant distance of 10 cm in field during *rabi*, 2016-17 at Research Farm, ICAR-IIWBR, Karnal. The parents, F<sub>1</sub> and F<sub>2</sub> generations derived from the crosses were screened under artificially created stripe rust epiphytotics. The planted materials were surrounded with infector lines to maintain heavy inoculum load and the materials were inoculated with the virulent barley stripe rust pathotype 57 (0S0). Randomly chosen F<sub>2</sub> plants were grown as F<sub>3</sub> progenies to validate the proposed hypothesis based on genetic analysis in F<sub>2</sub> generation during *rabi*, 2017-18.

Since the race 57 (0S0) is one of the most virulent pathotypes the pure inoculum of this race was considered for inoculating the parents and segregating generations. The infector rows were syringe inoculated at 21 days seedling stage (Zadok GS 20) with the inoculum of the uredospores of the stripe rust race 57

followed by repeated sprays of inoculum collected from the spreader rows on the genetic materials. Extra irrigation was given for maintaining cool and humid congenial micro-climate for optimum disease development. Stripe rust reactions were recorded from early to late flowering stages (Zadok GS 60-69), when spreader rows showed maximum susceptible reactions of 80S-100S. A modified Cobb scale (Peterson et al. 1948) was used to record adult plant reactions based on per cent infected host tissue and infection types (ITs). The different ITs were recorded as per the procedure described by Roelfs et al. (1992) i.e., R (resistant): necrotic areas with miniature uredinia; MR (moderately resistant): small uredinia surrounded by necrotic areas; MS (moderately susceptible): medium size uredinia and S (susceptible): large uredinia. Based on the adult plant reactions, the F<sub>2</sub> individuals were categorised into two groups merging resistant and moderately resistant plants (R+MR) and moderately susceptible and susceptible plants (MS+S). The Chi-Square test was employed to test the goodness of fit between observed and expected frequencies of resistant and susceptible plants in the segregating generations.

The barley genotypes, DWRB137 and DWRB143 showed resistance (0;) at seedling stage against barley stripe rust races 57, M and Q and adult plant (field) resistance (0) under artificial inoculation against with mixture of barley stripe rust races (Anonymous 2015). The F<sub>1</sub>s from Susceptible (S) × Resistant (R) crosses were found resistant at adult plant stage and thus showed dominant inheritance of resistance in the resistant parents. However, to determine the number of genes controlling resistance, the segregating generations (F<sub>2</sub> and F<sub>3</sub>) were raised and studied. In F<sub>2</sub> crosses (S × R) involving susceptible barley cultivar Lakhan with resistant parents DWRB137 and DWRB143, a total of 195 and 209 plants were categorized as resistant, respectively. Whereas, after merging resistant (R) and moderately resistant (MR) lts, a total of 265 and 254 plants were grouped as resistant for the crosses RD2035/DWRB137 and RD2035/DWRB143, respectively. Thus, in F<sub>2</sub> generations of the S × R crosses *viz.*, Lakhan/DWRB137, Lakhan/DWRB143, RD2035/DWRB137 and RD2035/DWRB143 segregation ratio of 3:1 was observed for resistant and susceptible plants (Table 1). This matched with the expected frequencies of resistant and susceptible plants, and suggested the presence of a single dominant gene each in the resistant parental stocks, DWRB137 and DWRB143.

Bakshi et al. (1964) and Luthra (1966) also reported dominant monogenic inheritance of stripe rust resistance in the barley genotype EB410.

Out of 68 F<sub>3</sub> progenies of the cross Lakhan/DWRB137, the 14 plant progenies were observed resistant and 15 were found susceptible, whereas 39 single plant progenies showed segregation for resistant and susceptible plants (Table 1). Similarly in the cross, RD2035/DWRB137, the 23 plant progenies were found resistant, 35 plant progenies as segregating, while 14

plant progenies showed susceptible reactions for stripe rust race 57 (0S0). Therefore, the F<sub>3</sub> segregation ratio of 1 (R):2 (Segregating):1(S) observed in all the four S × R crosses confirmed the monogenic dominant resistance in the genotypes DWRB137 and DWRB143 (Table 1). The test of allelism was carried out to determine the commonness or diversity of the resistance gene (s) between the resistant parents, DWRB137 and DWRB143. While all the F<sub>1</sub> plants of the cross DWRB137/DWRB143 showed stripe rust resistance under field conditions at the adult plant

**Table 1.** Observed and expected ratios in segregating generations for S × R and R × R crosses

Parent/cross	Ratio observed			Ratio expected			Segregation ratio	$\chi^2(C)$
	R	Seg.	S	R	Seg.	S		
<b>Cross I</b>								
Lakhan	0		All					
DWRB137	All		0					
F1 (Lakhan/DWRB137)	All		0					
F2 (Lakhan/DWRB137)	195		68	197		66	3R:1S	0.08
F3 (Lakhan/DWRB137)	14	39	15	17	34	17	1R:2Seg:1S	1.50
<b>Cross II</b>								
Lakhan	0		All					
DWRB143	All		0					
F1 (Lakhan/DWRB143)	All		0					
F2 (Lakhan/DWRB143)	209		75	213		71	3R:1S	0.30
F3 (Lakhan/DWRB143)	21	37	14	18	36	18	1R:2Seg:1S	1.42
<b>Cross III</b>								
RD2035	0		All					
DWRB137	All		0					
F1 (RD2035/DWRB137)	All		0					
F2 (RD2035/DWRB137)	265		96	271		90	3R:1S	0.53
F3 (RD2035/DWRB137)	23	35	14	18	36	18	1R:2Seg:1S	2.30
<b>Cross IV</b>								
RD2035	0		All					
DWRB143	All		0					
F1 (RD2035/DWRB143)	All		0					
F2 (RD2035/DWRB143)	254		81	251		84	3R:1S	0.14
F3 (RD2035/DWRB143)	19	28	18	16	33	16	1R:2Seg:1S	1.57
<b>Test of allelism</b>								
DWRB137	All		0					
DWRB143	All		0					
F1 (DWRB137/DWRB143)	All		0					
F2 (DWRB137/DWRB143)	321		12	313		21	15R:1S	4.06

$\chi^2(T) = 3.84$  &  $5.99$  at 1 & 2 d.f. at 5 % level of significance; R = resistant; Seg. = segregating; S = Susceptible and C = Calculated value

stage, the  $F_2$  populations segregated in the ratio of 15 resistant: 1 susceptible plants (Table 1) suggesting the presence of different genes in the two resistant parents. Therefore, these resistance sources should be studied further for determining the identity of their resistance genes. However, they can now be utilized for stripe rust resistance breeding in barley improvements programmes.

#### Author's contribution

Conceptualization of research (VK); Designing of experiments (VK, SCB); Contribution of materials (VK); Execution and coordination (VK, SCB, ASK, GPS), analysis of data (VK, GPS), compilation and interpretation (VK, SCB, GPS); Preparation of manuscript (VK).

#### Declaration

The authors declare no conflict of interest.

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