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POSTULATION AND VALIDATION OF LEAF RUST RESISTANCE GENES IN BREADWHEAT CULTIVARS HUW-12 AND WH-322

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ABSTRACT

On the basis of differential seedling reaction of HUW-12 and WH-322 against various brown rust races and comparing it with all the thirty Lr genes, the resistance genes present in these two varieties were postulated. Screening of F_2 seedlings derived from the crosses with susceptible Agra Local indicated the number of resistance genes present in these cultivars. The F_2 population of crosses WH-322 × Lr10 and HUW-12 × Lr14a failed to segregate. It is concluded that HUW-12 has Lr14a and WH-322 has Lr10, in addition to another unidentified Lr gene.

Key words: *Puccinia recondita tritici*, matching technique, Lr genes, *Triticum aestivum*.

Breeding rust resistant varieties is the only viable alternate to minimise losses due to the wheat rusts. And the use of specific genes for resistance has been the major approach [1]. However, this type of resistance is usually short lived and results in the boom and bust cycles on its overexploitation or when used indiscriminately. The span of usage of these specific resistance genes can be extended by proper management of the available resistance gene pool. But the difficulty lies in the lack of detailed information on the Lr genes operating in the Indian wheat varieties aganist brown rust (*Puccinia recondita* f. sp. *tritici*). The conventional genetic methods employed for the identification of resistance genes in cultivars involve more time and field space. Therefore, studies were undertaken to postulate the presence of probable brown rust resistance (Lr) genes in two breadwheat cultivars on the basis of infection types (IT) produced against different races of *P. recondita tritici* and proving the postulation so made through the test of allelism. Postulating the hypothetical genotypes based on IT data and validating through conventional genetic methods was first demonstrated by Loegering et al. [2]. Results on the identification of Lr genes present in the two breadwheat varieties, HUW-12 and WH-322 are presented below, on similar basis.

MATERIALS AND METHODS

The wheat cultivars HUW-12 and WH-322 were tested with all available races of leaf rust and reaction types were compared with that of all the 30 Lr genes. However, reaction types produced by 10 selected races, which gave differential reaction,

and near-isogenic lines with known Lr genes are presented in Table 1. Through the elimination procedure inference on the probable Lr gene(s), in these test cultivars was arrived at. The pedigree of the test cultivars added support in postulating the Lr gene(s). And the number and identity of these gene(s) were determined by analysing the F_2 population of a) cross with the universal susceptible variety Agra Local and b) cross with the near isogenic lines possessing the postulated gene.

In all cases seedling reaction was recorded 16 to 18 days after incubation at a mean maximum temperature of 22°C, in greenhouse, following [3]. Reaction types 0; to 2 were recorded as resistant and 3, 4 and X as susceptible. The goodness of fit (χ^2 test) for the genetic ratio were fitted. The probability level of the observed and expected proportion of the population gives an indication of the fitness of the postulations made.

RESULTS

Analysis of HUW–12. Seedlings of HUW–12 produced susceptible (4) IT to all the races of *P. recondita tritici* except against 11 (OR8), 63(OR8–1) and 106(OR9), where resistant (0; to 0;–1) IT was observed. Comparison of the IT data of HUW–12 and the lines with known Lr genes revealed a near similarity with Lr14a (Table 1). Therefore, it was postulated that HUW–12 possess only Lr14a out of the 30 Lr genes tested.

Table 1. Reaction types produced by test cultivars and some isogenic lines with known Lr genes against representative races of *Puccinia recondita* f. sp. *tritici*

Var./Lr line	Reaction against different races									
	11 (OR8)	12 (5R5)	63 (OR8-1)	77 (5R31)	77A (109R31)	77A-1 (109R23)	104 17R23)	106 (OR9)	162 (93R7)	162-A (93R15)
HUW 12*	0;	3-4	0;-1	4	4	4	2-3	0;	4	4
WH 322**	0;	0;	0;	0;	4	4	0;-1	0;	0;	0;
Lr1	0;	0;	0;	4	4	4	4	0;	0;	0;
Lr2a	0;	0;-1	0;	4	4	4	1-3	0;-1	4	4
Lr3	0;	4	0;	4	4	4	4	0;	4	4
Lr10	0;	0;-2	0;	0;	4	4	0;-1	0;	2-3	2-3
Lr14a	0;	4	0;	4	4	4	4	0;	4	4
Lr15	0;	0;	0;	4	4	4	0;	0;	0;	0;
Lr18	0;-2	2-3	0;	2-3	4	2-3	0;-2	0;	4	4
Lr20	4	0;-1	4	4	4	0;-1	0;-1	2-3	0;	4
Lr24	0;	0;	0;	0;	0;	0;	0;	0;	0;	0;

* Reaction types similar to Lr14a. ** Reaction types nearly similar to Lr10.

In the F_2 population derived from the cross with Agra Local, out of 150 seedlings, 117 were resistant and 33 susceptible. This segregation pattern of F_2 population fits well with the monogenic nature of resistance, i.e. 3 (resistant) : 1 (susceptible). The χ^2 value (0.71) and high probability level (P=0.3–0.5), further

confirm that resistance to *P. recondita tritici* in this cultivar is governed by a single gene (Table 2).

Also all the 144 F_2 seedlings of the cross with the tester near-isogenic line Lr14a were resistant to race 106 (OR9). Lack of segregation in the test of allelism shows that a single Lr gene operates in HUW-12, i.e. Lr14a.

Analysis of WH-322. In the seedling stage, WH-322 showed susceptible (4) IT only against races 77A(109R31) and 77A-1(109R23). This cultivar was resistant against all other brown rust races that were tested. Comparison of IT lines having known Lr genes with those produced by WH-322 showed near similarity of IT with Lr10, except to races 162(93R7) and 162A(93R15). The near-isogenic lines for Lr10 produced 2-3 IT against these races, whereas WH-322 showed resistant (0) IT (Table 1). Therefore, it was postulated that WH-322 possess two Lr genes, of which one is Lr10 and the other gene remains unidentified so far.

Of the 96 F_2 seedlings derived by crossing with Agra Local, 88 were resistant and 8 susceptible. The segregation pattern of F_2 population fitted in a 15 (resistant) : 1 (susceptible) ratio, indicating duplicate nature of the resistance genes operating against race 104(17R23) of brown rust. The 1.46 χ^2 value and P=0.2-0.3 (Table 2) also confirm the presence of two Lr genes in WH-322. All the 104 F_2 seedlings of the cross with Lr10 near-isogenic line were resistant, and the absence of segregation confirms the identity of Lr10 in WH-322.

Table 2. Segregation pattern of F_2 seedlings against specific races of *Puccinia recondita* f. sp. *tritici*

Cross	Race	Resistant	Susceptible	Total
HUW 12 × Agra Local	106(OR9			
Observed		117	33	150
Expected (3:1)		111	37	148
	$\chi 2$=0.71	P=0.50-0.30		
HUW 12 × Lr14a	106(OR9)	144	0	144
WH 322 × Agra local	104(17R23)			
Observed		88	8	96
Expected (15:1)		90	6	96
	$\chi 2$=1.46	P=0.30-0.20		
WH 322 × Lr10	77(45R31)	104	0	104

DISCUSSION

The effectiveness of IT data has been amply demonstrated for postulating the hypothetical genotypes in the host cultivars [2, 4-8]. Based on seedling reaction of the test cultivars against individual races, postulation on the probable Lr genes present, was made. These postulations were further validated on the basis of the information collected through genetical studies.

The gene Lr14a is loosely linked with gene Sr17 having a crossing over frequency of 18%. It is also reported to have 20% recombination with a powdery mildew resistance gene Pm5 [9]. Although Lr14a is susceptible to most of the brown rust races in India but can be useful for achieving multiple disease resistance because of its linkage with Sr17 and Pm5.

Mayo 54 and Gabo are involved in the development of WH–322 (HD 2216 × HD 2009). Presence of Lr10 in both Mayo 54 [10] and Gabo [9] has been reported. It is likely that Lr10 might have come in WH–322 either from Mayo 54 or Gabo. The second Lr gene in WH–322 could not be identified on the basis of IT data generated with the available races of *P. recondita tritici*. But based on the F_2 reaction to race 104(17R23) giving a 15:1 (R:S) segregation, the unidentified gene in WH–322 could be only one, different from Lr2a, Lr3, Lr13, Lr17 and Lr18. Despite the inherent limitation of the matching technique it is still quite useful in the identification of the probable resistance genes in the cultivars. This procedure permits the evaluation of a large number of host cultivars in much less time and space. Moreover, postulations so made can be confirmed by the test of allelism.

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