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INHERITANCE OF STEM RUST RESISTANCE IN A WHEAT-RYE RECOMBINANT LINE 'SELECTION 212'

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ABSTRACT

Selection 212, a wheat-rye recombinant developed through homoeologous recombination between wheat and rye chromosomes using monosomic 5B of variety Chinese Spring was studied for inheritance of resistance against stem rust pathogen (*Puccinia graminis* f. sp. *tritici*). The F_1, the F_2, the F_3, the BC_1F_1 and the BC_1F_2 generations of the crosses involving 'Selection 212' and two susceptible lines Agra Local and Chinese Spring were tested in seedlings with pathotypes 122 and 40A of *P. graminis tritici*. A single recessive gene that controlled resistance to both the pathotypes was determined. Correlated behaviour of the F_2 backcross families of the cross (Sel.212 × AL) × AL with both the pathotypes revealed that the same resistance gene is providing resistance to pathotypes 122 and 40A. In addition, an adult plant resistance gene *Sr2* was also identified.

Key Words : Wheat, rye (*Secale cereale*), stem rust (*Puccinia graminis*), inheritance, recessive

Stem rust of wheat caused by *Puccinia graminis* Pres. f.sp. *tritici* Eriks. and Henn. is the most devastating disease of wheat crop in warmer climates. Breeding for rust resistance is the most feasible and practical approach to check losses caused by rust diseases. The evolution of new pathotypes on widely grown resistant cultivars necessitates identification of new sources of resistance for continuous process of resistance breeding. In an effort 'Selection 212', (Sel. 212) a line with new source of resistance was developed by homoeologous chromosome recombination between wheat and rye using monosomic 5B of variety Chinese Spring [1]. Sel. 212 when tested at seedling stage with 20 pathotypes of *P. graminis tritici* was found resistant to all the pathotypes [2]. The present study reports the inheritance of resistance to stem rust at seedling and adult plant growth stages using pathotypes 122 and 40A which identify resistance in Sel. 212, transferred from rye.

MATERIALS AND METHODS

The F_1, F_2, F_3, BC_1F_1 and BC_1F_2 generations of the crosses involving Sel. 212 and susceptible lines Agra Local (AL) and Chinese Spring (CS) along with parental line Sonalika were tested with pathotypes 122 and 40A of *P. graminis tritici* at seedling stage. Adult plant tests were, however, conducted with only 40A a highly virulent and predominant pathotype. Avirulence/virulence formulae of these pathotypes are as follows.

Pathotype 122 = *Sr8a, 9e, 24, 28, 30, 37/Sr5, 7b, 9b, 11, 13, 18, 19, 20, 21*

Pathotype 40A = *Sr13, 21, 24, 30, 37/Sr5, 7b, 8a, 9b, 9e, 11, 18, 19, 20, 28*

Both these pathotypes are avirulent on *Sr25, Sr26, Sr27, Sr31, Sr32* and virulent on *Sr6, Sr9a, Sr9d, Sr9f and Sr16.*

Seedling tests : Seedlings were grown in rectangular trays (11" × 4" × 3"). Ten day old seedlings were inoculated with pure urediospore cultures using lanceolate needle and incubated in humidity chambers for 48h. Thereafter, seedlings were shifted to glass house benches. Reaction types were recorded according to the scale described by Stakman *et al.* [3].

Table 1. Seedling and adult plant reaction on parental lines and F_1s involving Sel. 212 against pathotypes 122 and 40A of *Puccinia graminis* f. sp. *tritici*

Parent\ Cross	Rust reaction					
	Seedling				Adult plant	
	122		40A		40A	
	Infection type	No. of seedlings	Infection type	No. seedlings	Field response	No. of plants
Selection 212	1+ (R)	17	12= (R)	16	TR (R)	19
Agra Local (AL)	4 (S)	20	4 (S)	18	80S (S)	17
Chinese Spring (CS)	4 (S)	16	4 (S)	19	90S (S)	14
Sonalika	3+ (S)	18	3+ (S)	15	60MS (R)	11
F_1 (Sel. 212 × AL)	4 (S)	10	3+ (S)	08	80S (S)	06
F_1 (Sel. 212 × CS)	3+ (S)	09	3+ (S)	09	80S (S)	07
F_1 (CS × Sel. 212)	3+ (S)	06	3+ (S)	06	NT	

NT = Not tested, R = Resistant, S = Susceptible

Adult plant tests

The material were grown in one metre long rows in an isolated nursery. Spreader row was planted at regular intervals and also all around the nursery for uniform spread of rust infection. Spreader rows were inoculated at boot leaf stage with urediospore suspension in water with the help of hypodermal syringe. Rust severity was recorded according to the scale described by Peterson *et al.* [4].

RESULTS AND DISCUSSION

Results of the tests on seedlings and adult plants of parental lines and F_1s involving Sel.212 against pathotypes 122 and 40A are presented in Table 1. Sel. 212 was resistant both at seedling and adult plant stages. Sonalika, the parental line of Sel. 212 was susceptible at seedling stage but showed a low degree of resistance at adult plant stage. All the F_1s of crosses Sel.212 × AL, Sel.212 × CS and the reciprocal cross CS × Sel. 212 were susceptible at seedlings and adult plants to both the pathotypes showing that resistance in Sel. 212 to pathotype 122 and 40A is recessive.

Results of the tests on F_2, F_3 and BC_1F_1 generations involving Sel. 212, at seedling and adult plant stages against pathotypes 122 and 40A are presented in Table 2. When tested with pathotype 122 the F_2 segregation of 120 seedlings of cross Sel. 212 × AL gave a close fit to 1R:3S ratio ($\chi^2 = 0.044$, $P > 0.80$). The F_2 segregation of the cross Sel. 212 × CS also conformed to 1R:3S ratio. A similar segregation pattern of the reciprocal cross (CS × Sel. 212) indicated the absence of cytoplasmic effect. This single recessive gene for resistance in the F_2 was confirmed by results obtained from the backcrosses (Sel. 212 × AL) × Sel. 212 and (Sel. 212 × CS) × Sel. 212 which showed monogenic segregation of 1R:1S without heterogeneity among the crosses ($\chi^2 = 0.003$, $P > 0.95$).

In tests against the pathotype 40A at seedling stage' F_2s of the crosses Sel. 212 × AL, Sel. 212 × CS and the reciprocal cross CS × Sel. 212 segregated into 1R : 3S ratio without significant heterogeneity among the crosses suggesting one recessive gene for resistance. Confirmation of F_2 results was obtained from the F_3 of Sel. 212 × AL, BC_1F_1s of (Sel. 212 × AL) × Sel. 212 and (Sel. 212 × CS) × Sel. 212. The distribution of the 28 backcross F_2 families of the cross (Sel. 212 × AL) × AL against pathotypes 122 and 40A into 15 segregating and 13 susceptible fits well into 1 segregation : 1 true breeding susceptible segregation pattern. Intra-family segregation into 1R : 3S ratio of segregating BC_1F_2 families gave further support to the findings reported above. Correlated behaviour of the F_2 families of the backcross (Sel.212 × AL) × AL with pathotypes 40A and 122 is presented in Table 3. Out of the 28 BC_1F_2

Table 2. Seedling and adult plant reaction on F_2, F_3 and BC_1F_1 generations involving Sel. 212 against pathotypes 122 and 40A of *Puccinia graminis* f. sp. *tritici*

Cross	No. of seedlings/plants/families				Expected ratio R:S/ R:S:Seg	χ^2	P value
	Total	Res.	Sus.	Seg.			
Seedling reaction for Pathotype 122							
F_2 (Sel. 212 × AL)	120	31	89	0	1:3	0.044	0.90-0.80
F_2 (Sel. 212 × CS)	119	23	96	0	1:3	2.042	0.20-0.10
F_2 (CS × Sel. 212)	244	73	171	0	1:3	3.146	0.10-0.05
Total of 3 crosses	483	127	356	0	1:3	0.431	0.70-0.50
			Heterogeneity χ^2			4.801	0.10-0.05
BC_1F_1 [(Sel. 212 × AL) × AL]	77	0	77	0	0:1		
BC_1F_1 [(Sel. 212 × AL) × Sel. 212]	41	24	17	0	1:1	1.195	0.30-0.20
BC_1F_1 [(Sel. 212 × CS) × Sel. 212]	38	22	16	0	1:1	0.947	0.50-0.30
Total of 2 crosses	79	46	33	0	1:1	2.139	0.20-0.10
			Heterogeneity χ^2			0.003	0.98-0.95
Seedling reaction for pathotype 40A							
F_2 (Sel. 212 × AL)	93	23	70	0	1:3	0.003	0.98-0.95
F_2 (Sel. 212 × CS)	185	54	131	0	1:3	1.731	0.20-0.10
F_2 (CS × Sel. 212)	194	43	151	0	1:3	0.832	0.50-0.30
Total of 3 crosses	472	120	352	0	1:3	0.045	0.90-0.80
			Heterogeneity χ^2			2.521	0.30-0.20
F_3 (Sel. 212 × AL)	80	26	12	42	1:1:2	5.100	0.10-0.05
BC_1F_1 [(Sel. 212 × AL) × AL]	42	0	42	0	0:1		
BC_1F_1 [(Sel. 212 × AL) × Sel. 212]	34	20	14	0	1:1	1.059	0.50-0.30
BC_1F_1 [(Sel. 212 × CS) × Sel. 212]	26	15	11	0	1:1	0.615	0.50-0.30
Total of 2 crosses	60	35	25	0	1:1	1.667	0.20-0.10
			Heterogeneity χ^2			0.007	0.95-0.90
Adult plant reaction to pathotype 40A							
F_2 (Sel. 212 × AL)	130	56	74	0	7:9	0.024	0.90-0.80
F_2 (Sel. 212 × CS)	106	51	55	0	7:9	0.820	0.50-0.30
Total of 2 crosses	236	107	129	0	7:9	0.242	0.70-0.50
			Heterogeneity χ^2			0.602	0.50-0.30
F_3 (Sel. 212 × AL)	89	40	07	42	7:1:8	0.541	0.80-0.70
BC_1F_1 [(Sel. 212 × AL) × AL]	28	0	28	0	0:1		
BC_1F_1 [(Sel. 212 × AL) × Sel. 212]	35	27	08	0	3:1	0.086	0.80-0.70

families, 15 segregated for resistance while 13 families were susceptible to both 40A and 122. Chi-square linkage was highly significant. The close association for resistance and a similar type of infection against both 40A and 122 indicated the operation of same gene for resistance to both the pathotypes.

Table 3. **Correlated behaviour of F_2 families of the backcross (Sel. 212 × AL) × AL when tested with pathotypes 40A and 122 of *P. graminis tritici* at seedling stage**

	Joint segregation of families				
		Pathotype 40A			
		Seg.	Sus.	Total	$\chi^2_{1:1}$= 0.143, 1 df, P>0.70 for both the pathotypes 40A and 122
Pathotype 122	Seg.	15	0	15	
	Sus.	0	13	13	
	Total	15	13	28	

$\chi^2_{(Compound)1:1:1:1}$ = 28.286, 3 df, P < 0.001; $\chi^2_{difference(Linkage)}$ = 28.0, 1 df, P < 0.001

Adult plant responses on different filial generations of crosses involving Sel. 212 to pathotype 40A under field conditions are also presented in Table 2. The F_2 of these crosses segregated into 7R:9S ratio suggesting that the resistance in Sel. 212 is recessive and digenically controlled. Adult plant responses for resistant F_2 segregants ranged from TR to 50MS. Twelve out of 130 plants showed a similar reaction as observed on parental line (TR). It conforms to the proportion of 1/16 (χ^2 = 1.549, P>0.10), an expected segregation value for carrying two recessive genes in homozygous condition. The high degree of resistance in Sel. 212 could be due to interaction of two recessive genes for resistance. The digenic control of resistance by recessive genes was confirmed by distribution of F_3 families into 7R : 8Seg: 1S classes (χ^2 = 0.541, P>0.70) and segregation of BC_1F_1 plants of cross (Sel.212 × AL) × Sel. 212 into 3R : 1S ratio. As expected all the BC_1F_1 of cross (Sel. 212 × AL) × AL were susceptible as both the genes for resistance are recessive.

Seedling tests with pathotype 40A revealed the presence of one recessive gene while adult plant tests suggested the operation of two recessive genes for resistance in Sel. 212. It suggests that apart from seedling resistance gene which is effective throughout the plant life, an adult plant resistance gene which becomes effective only at adult stage of plant growth is operating in Sel. 212. Based on the presence of pseudo-black chaff and chlorotic seedlings at high temperature in Sel.212, the two

Table 4. Inheritance of leaf chlorosis, associated with *Sr2*, in the F_1 and F_2 of the cross Sel. 212 × Chinese Spring and the parental lines at a mean temperature range of 16°-38°C

Parent/Cross	Population	No. of seedlings			$\chi^2_{(1:3)}$	P value
		Chlorosis present	Chlorosis absent	Total		
Selection 212	P_1	16	0	16		
Chinese Spring (CS)	P_2	0	19	19		
Sonalika (control)		18	0	18		
Sel. 212 × CS	F_1	0	05	05		
Sel. 212 × CS	F_2	78	188	266	2.652	0.20-0.10

established markers of *Sr2*, this adult plant resistance gene was postulated to be *Sr2* [5, 6]. This gene could have been derived from Sonalika [7, 8], one of the parent of Sel. 212, which also showed pseudo-black chaff and seedling chlorosis.

The presence of *Sr2* which otherwise is not expressed in seedling stage can be identified with the help of seedling chlorosis. Inheritance of leaf chlorosis associated with *Sr2* was studied in cross Sel. 212 × CS (Table 4). Sel. 212 seedlings were chlorotic while seedlings of Chinese Spring did not show any chlorosis. Sonalika, a known carrier of *Sr2* was chlorotic. The F_1 seedlings of cross Sel. 212 × CS were non-chlorotic suggesting that leaf chlorosis is recessive. The F_2 seedlings segregated for chlorosis and non-chlorosis in 1:3 ratio revealing the recessive monogenic segregation of this trait. The mode of inheritance of *Sr2* is also recessive [5]. Thus it was concluded that Sel.212 possesses an adult plant resistance gene (*Sr2*) and a seedling undescribed recessive gene for resistance to stem rust.

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