

Genetics of stem rust resistance in four popular durum wheat cultivars

A. Divya^{*}, T. L. Prakasha, S. Chand¹, A. N. Mishra, V. G. Dubey, Prakash Malviya, Pramod Prasad² and S. V. Sai Prasad

ICAR-Indian Agricultural Research Institute, Regional Station, Indore, M.P.; ¹School of Life Sciences, Devi Ahilya Vishwavidyalaya, Indore, M.P.; ²ICAR-IIWBR, Regional Station, Flowerdale, Shimla

(Received: December 2019; Revised: April 2020; Accepted: April 2020)

Abstract

A study was conducted to understand the mode of inheritance and extent of diversity of stem rust resistance in four popular durum wheat cultivars of central India viz., HI 8498 (Malav Shakti), HI 8663 (Poshan), HI 8713 (Pusa Mangal) and HI 8737 (Pusa Anmol) using Puccinia graminis tritici (Pgt) pathotypes 15-1 (123G15) and 40-3 (127G29). These cultivars were crossed with susceptible parents *i.e.*, Motia and Malvi Local and were also crossed among themselves in half diallel fashion. The F₂ and F₃ segregation data revealed that a single dominant gene each controlled resistance to the pathotype 40-3 in HI 8713 and HI 8663, while two dominant genes each governed resistance to this pathotype in HI 8737 and HI 8498. A single dominant gene each conditioned resistance to the pathotype 15-1 in all the four cultivars. The F₂ segregation data of the intercrosses among the resistant parents showed that three different resistance genes controlled resistance among four cultivars against each Pgt pathotype 40-3 and 15-1. These genes seem to be different from the most commonly postulated stem rust resistance genes in Indian durum wheat germplasm viz., Sr11, Sr12, Sr7b and Sr9e which are ineffective/less effective against the test pathotypes. Hence, the genes identified in the present study can be utilized in broadening the genetic base of stem rust resistance in Indian durum wheat.

Key words: Durum wheat, pathotype, resistance and stem rust

Introduction

Durum wheat (*Triticum turgidum* ssp. *durum* L.) is the second most important wheat species, occupying nearly 10% of the wheat area in India with 2.5 million tons of annual production. It has tremendous export potential due to its increasing global demand, better quality, value addition potential and better price in

international market (Gautam et al. 2013). Durum is the hardest of all wheats and predominantly grown in central and peninsular zones of India.

Wheat stem rust (Puccinia graminis f. sp. tritici) is historically the most damaging disease of wheat leading to extensive yield losses. Until 1980s, durum wheat cultivation in Central India was on the verge of extinction due to low yield potential and susceptibility to rust diseases (Nagarajan and Joshi 1975; Joshi et al. 1980). However, development and deployment of high yielding and rust resistant varieties brought durum wheat back in cultivation in central India. Recently released durum wheat varieties viz., HI 8498, HI 8663, HI 8713, HI 8737, HI 8759 and HI 8777 have high vield potential along with rust resistance and tolerance to drought and heat ensuring high profitability to the farmers. Durum wheat cultivation is a scientific necessity as its spectrum of rust resistance differs from that of the bread wheat varieties. The aforesaid new durum varieties are generally field resistant to prevalent and bread wheat virulent rust pathotypes including race 77-group of leaf rust and 40A and 40-1 of stem rust (Anonymous 2017). However, the evolution of novel rust races and their variants can occur through recombination of their genetic material, migration, mutation followed by selection.

Hence, use of cultivars with diverse resistance genes is the best method for achieving adequate and long lasting genetic control of diseases affecting wheat, including rusts (Pink 2002). Therefore, identification of resistance genes and determining the extent of their diversity among the existing wheat cultivars or wheat

*Corresponding author's e-mail: divya.ambati@icar.gov.in

Published by the Indian Society of Genetics & Plant Breeding, A-Block, F2, First Floor, NASC Complex, IARI P.O., Pusa Campus, New Delhi 110 012; Online management by www.isgpb.org; indianjournals.com

relatives' species can contribute to the effective management of wheat rusts (Kolmer 2003 and Sumikova and Hanzalova 2010). However, information is limited on the nature and the genetic basis of rust resistance in durum wheat. Hence, a study was planned during 2015-18 to gain an insight into the inheritance and extent of diversity of stem rust resistance in four popular durum wheat varieties.

Materials and methods

The popular resistant (R) durum wheat cultivars (R parents) *viz.*, HI 8498 (Malav Shakti), HI 8663 (Poshan), HI 8713 (Pusa Mangal) and HI 8737 (Pusa Anmol) were crossed with susceptible (S) female parental lines (S parents) *viz.*, Malvi Local and Motia. The parentage of these test cultivars is as follows:

Variety	Parentage
HI 8737	HI 8177 / HI 8158// HI 8498
HI 8713	HD 4672 / PDW 233
HI 8663	HI 8177 / HI 8185
HI 8498	CR "S -GS S /A-9-30-1// RAJ 911
Malvi Local	Landrace
Motia	Landrace

The resistant cultivars were intercrossed in all possible combinations without the reciprocals to obtain F_1 s. Space-sown F_1 plants were harvested individually to obtain F_2 progeny. All the parents, F_1 s and F_2 plants were screened in glasshouse for two selected stem rust (*Pgt*) pathotypes *viz.*, 40-3(127G29) and 15-1(123G15). The avirulence/virulence formula of these pathotypes is as given in Table 1 (Jain et al. 2013).

In the glasshouse tests, the parents and F_2 populations of SxR and RxR crosses were inoculated at the seedling (two-leaf) stage by spraying uredospores of the test pathotypes. Plants were placed

in a dew chamber overnight to ensure spore germination and completion of the infection process and then transferred to glass house benches kept at 18 to 25°C. Infection types displayed on the leaves were recorded about 10-15 days after inoculation according to 0 to 4 scale (Mishra et al. 2015). The F_3 families were grown from random F_2 plants of susceptible parent x resistant parent (SxR) crosses to confirm the results of F_2 ratios. The F_3 families of SxR crosses were screened along with parents with two selected stem rust (*Pgt*) pathotypes to confirm the F_2 ratios. The F_3 families were grouped into homozygous resistant, segregating and homozygous susceptible populations.

Pheno-typic frequencies of observed and expected ratios of resistant and susceptible plants were tested for goodness-of-fit using chi-square test. The crosses of test cultivars with susceptible lines were studied to determine the number of genes controlling resistance, whereas intercrosses among resistant cultivars were studied to determine the extent of diversity for rust resistance genes. All the six parents along with the lines carrying known *Sr* genes reported in Indian durum wheat germplasm were seedling tested with both the *Pgt* pathotypes in the glass house to gain an insight and to the identity of the genes conferring resistance in the test cultivars.

Results

Seedling resistance to Puccinia graminis f. sp. tritici pathotype 40-3

The genetic analysis of the ratio of resistant and susceptible plants in F_2 populations of the susceptible x resistant crosses (Table 2) indicated that two independent dominant genes each conditioned resistance in HI 8498 (15R:1S, P=0.82) and HI 8737 (15R:1S, P=0.21-0.30); whereas a single dominant gene governed resistance in HI 8713 (3R:1S, P=0.38-0.72) and HI 8663 (3R:1S, P = 0.62). The susceptible

 Table 1.
 A list of arivulant and virulant gene against stem rust pathotypes 15-1 and 40-3

Pathotype	Avirulent to Sr genes	Virulent to Sr genes
123G15 (15-1)	Sr7a, Sr11, Sr24, Sr25, Sr26, Sr27, Sr31, Sr32, Sr33, Sr35, Sr37, Sr39, Sr40, Sr43 SrGt, SrTmp, SrTt3	Sr2, Sr5, Sr6, Sr7b, Sr8a, Sr8b, Sr9a, Sr9b, Sr9d, Sr9e, Sr9f, Sr9g, Sr10, Sr12, Sr13, Sr14, Sr15, Sr16, Sr17, Sr18, Sr19, Sr20, Sr21, Sr22, Sr23, Sr28, Sr29, Sr30, Sr34, Sr36, Sr38, Sr42, Sr44, SrMcN, SrWld
127G29 (40-3)	Sr21, Sr22, Sr24, Sr25, Sr26, Sr27, Sr31, Sr32, Sr33, Sr35, Sr36, Sr37, Sr39, Sr40, Sr42, Sr43, SrTmp, SrTt3	Sr2, Sr5, Sr6, Sr7a, Sr7b, Sr8a, Sr8b, Sr9a, Sr9b, Sr9d, Sr9e, Sr9f, Sr9g, Sr10, Sr11, Sr14, Sr15, Sr16, Sr17, Sr18, Sr19, Sr20, Sr23, Sr28, Sr29, Sr30, Sr34, Sr38, Sr44, Sr McN, SrGt

Crosses	ses Number of F ₂ plants $\chi 2$ P Number of F ₃ families		amilies	χ 2	Р				
	R	S			HR	SEG	HS		
Malvi local / HI 8498	25	30			34	40	6	0.23 (7:8:1)	0.89
Malvi local / HI 8663	62	9			17	26	16	0.86 (1:2:1)	0.64
Malvi local / HI 8713	50	15	0.12 (3:1)	0.72	24	35	20	1.43 (1:2:1)	0.49
Malvi local / HI 8737	82	08	1.07 (15:1)	0.30	42	35	3	2.82 (7:8:1)	0.29
Motia / HI 8498	99	6	0.05 (15:1)	0.82	41	35	3	2.47 (7:8:1)	0.29
Motia / HI 8663	64	24	0.24 (3:1)	0.62	15	30	9	2.06 (1:2:1)	0.36
Motia / HI 8713	88	24	0.76 (3:1)	0.38	15	28	13	0.14 (1:2:1)	0.93
Motia / HI 8737	100	10	1.51 (15:1)	0.21	37	38	5	0.21 (7:8:1)	0.90

Table 2. Segregation for Puccinia graminis tritici pathotype 40-3 in F2 populations and F3 families of SxR crosses

 $F_2 = R$, resistant; S = susceptible; $F_3 = HR$, homozygous resistant; SEG = segregating; HS = homozygous susceptible. Values for significance at P = 0.05 are 3.841 for 1 d.f.

plants in F₂ populations of SxR crosses *viz.*, Malvi local/HI 8498 and Malvi Local/HI 8663 did not fit the 3R:1S ratio. However the F₃ families of these crosses segregated in 7HR:8SEG:1HS and 1HR:2SEG:1S ratios, respectively which confirmed the presence of two dominant and a single dominant gene in HI 8498 and HI 8663, respectively.

The F_2 ratios of RxR crosses (allelic tests) *viz.*, HI 8713/HI 8498 and HI 8713/HI 8663 showed segregation (Table 3), indicating that the resistance

Table 3.	Segregation	for	Puccinia	graminis	tritici
	pathotype 40-3	3 in F	2 population	ns of RxR cr	osses

Crosses	Ν	lo. of F ₂	2 pla	ints ₂ 2	Р
	_	R	S		
HI 8713 / H	H 8498	839	98	28.32(15:1) 1.02363E-07
HI 8713 / H	HI 8663	1247	82	0.01(15:1)	0.90
HI 8713 / H	HI 8737	1058	0	70.53(15:1) 4.52555E-17
HI 8498 / H	HI 8663	1061	0	70.73(15:1) 4.08925E-17
HI 8498 / H	HI 8737	919	0	61.26(15:1) 4.98448E-15
HI 8663 / H	HI 8737	1189	0	79.26(15:1) 5.42676E-19

 F_2 : R = Resistant; S = Susceptible; Values for significance at P = 0.05 are 3.841 for 1 d.f.

genes among HI 8498 and HI 8663 are diverse. There were no susceptible plants in the other four crosses indicating that at least one gene was common among them.

Seedling resistance to Puccinia graminis f. sp. tritici pathotype 15-1

The analysis of F_2 populations' segregation data (Table 4) of the susceptible x resistant crosses indicated the

presence of single dominant gene each in all the test cultivars (3R:1S with P value ranging from 0.06-0.69). The segregation ratio of F_3 families (1HR:2SEG:1HS) of these crosses (Table 4) also confirmed the F_2 segregation data with P value ranging from 0.34-0.98. The F_2 ratios of all RxR crosses (Table 5) showed segregation for susceptible plants, except HI 8713/HI 8737 indicating a total of three dominant genes controlling resistance among the four test cultivars, with a gene being common in HI 8713 and HI 8737.

Discussion

The genetic analysis of F2 populations of the susceptible x resistant crosses screened for indicated that two independent dominant genes each conditioned resistance in HI 8498 and HI 8737 and a single dominant gene governed resistance in HI 8713 and HI 8663. The allelic tests of these parental lines showed presence of three diverse resistance genes among the four cultivars and at least one diverse gene for resistance in HI 8498 and HI 8663 and at least one common resistance gene between the cultivars HI 8713 and HI 8737. The analysis of F₂ populations' segregation data of the susceptible x resistant crosses indicated the presence of single dominant gene each in all the four test cultivars. This study revealed that resistance in each of the four cultivars against the Pgt pathotypes 15-1 and 40-3 is controlled by a single dominant or two independent dominant genes. The rust resistance in wheat being controlled by one or two genes was reported in many studies (Zia UI Qamar et al. 2009; Olivera et al. 2012; Mishra et al. 2015). There is commonness for at least one Sr gene conferring resistance against the two test pathotypes between HI 8737 and HI 8713.

Crosses	Number of F ₂ plants		χ2	Р	Number of F ₃ families			χ 2	Р
	R	S			HR	SEG	HS		
Malvi local / HI 8498	41	10	0.79 (3:1)	0.37	28	30	20	5.79	0.05
Malvi local / HI 8663	36	19	2.67 (3:1)	0.10	22	34	15	1.54	0.47
Malvi local / HI 8713	41	12	0.15 (3:1)	0.69	20	26	15	2.14	0.34
Malvi local / HI 8737	75	21	0.50 (3:1)	0.47	20	32	15	0.88	0.64
Motia / HI 8498	62	25	0.64 (3:1)	0.42	20	27	17	1.84	0.39
Motia / HI 8663	72	14	3.48 (3:1)	0.06	20	30	13	1.69	0.42
Motia / HI 8713	57	25	1.31 (3:1)	0.25	25	48	28	0.04	0.98
Motia / HI 8737	63	16	0.94 (3:1)	0.32	32	41	19	4.76	0.06

Table 4. Segregation for Puccinia graminis tritici pathotype 15-1 in F₂ populations and F₃ families of SxR crosses

F2 = R, resistant; S = susceptible; F3 = HR, homozygous resistant; SEG = segregating; HS = homozygous susceptible. Values for significance at P = 0.05 are 3.841 for 1 d.f.

Table 5.	Segregation for Puccinia graminis tritici
	pathotype 15-1 in F ₂ populations of RxR crosses

Crosses	No. of F	₂ pla	Р		
	R	S			
HI 8713 / HI 8498	866	46	2.26(15:1)	0.13	
HI 8713 / HI 8663	3 1066	33	19.7(15:1)	8.69762E-06	
HI 8713 / HI 8737	967	0	64.46(15:1)	9.81793E-16	
HI 8498 / HI 8663	3 997	24	26.49(15:1)	2.64238E-07	
HI 8498 / HI 8737	7 897	53	0.73(15:1)	0.39	
HI 8663 / HI 8737	7 1134	86	1.33(15:1)	0.24	

F2: R = resistant; S = susceptible; Values for significance at P = 0.05 are 3.841 for 1 d.f.

Among the 57 Sr genes identified till now, Sr genes reported in tetraploid wheat include Sr2, Sr9d, Sr9e, Sr9g, Sr11, Sr12, Sr13, Sr14, and Sr17 (McIntosh et al. 1995, 2013). Association mapping of 183 diverse durum wheat accessions revealed the presence of Sr13, and suggested likely contributions of Sr9h, Sr14, Sr17 and Sr28 in providing resistance to Ug99 and durum-specific Ethiopian Pgt races (Letta et al. 2013). The seedlings of the parents and lines carrying known stem rust resistance genes such as Sr7a, Sr7b, Sr8a, Sr8b, Sr9b, Sr9e, Sr9f, Sr9g, Sr11, Sr12, Sr13, Sr14, Sr17 and Sr23 were tested with both pgt pathotypes 40-3 and 15-1 in the glass house. Seedling reactions of the parents didn't match with any of the lines with known Sr lines tested with the two pathotypes (Table 6). Hence, the genes identified conferring stem rust resistance in the test cultivars seem to be different from Sr7b, Sr9g, Sr9e, Sr11, Sr12, Sr13, Sr14, and Sr17 as these genes were found to

Table 6.Seedling infection types of the four cultivars and
the genes of interest tested with Pgt pathotypes
40-3 and 15-1

Variety	40-3	15-1	Genes	40-3	15-1
HI 8498	2 ne	;1+	Sr12	4	34
HI 8663	11+	;1+	Sr8a	34	34
HI 8713	12	;1+ch	Sr8b	34	34
HI 8737	12	;1+	Sr9f	4	34
Sr7a	34	34	Sr9g	4	23+
Sr7b	34	34	Sr13	34	34
Sr9b	2+	34	Sr14	34	34
Sr9e	4	34	Sr17	4	34
Sr11	4	23+	Sr23	34	34

be ineffective/less effective against the pathotypes 40-3 and 15-1 in the seedling tests.

The avirulence/virulence formula of two *pgt* pathotypes shows that these are virulent to two other *Sr* genes reported in durum background *viz., Sr9d* and *Sr28* which rules out the presence of these resistance genes in the test cultivars. The two test pathotypes 15-1 and 40-3 are avirulent to other *Sr* genes *viz., Sr24, Sr25, Sr26, Sr27, Sr31, Sr32, Sr33, Sr35, Sr37, Sr39, Sr40, Sr43 SrGt, SrTmp, SrTt3* based on the virulence/avirulence formula of these pathotypes. However, these *Sr* genes have not been reported in durum germplasm worldwide till now.

Thus, results of our study showed that the genes identified in the four test cultivars conferring resistance to both the *Pgt* pathotypes 40-3 and 15-1 seem to be

different from commonly occurring Sr genes in durum germplasm reported globally till now. Hence, these cultivars hold promise as additional sources of stem rust resistance in durum wheat and could be effectively utilized in widening the stem rust resistance base of durum wheat.

Authors' contribution

Conceptualization of research (AD, ANM, SC); Designing of the experiments (AD, ANM, SC); Contribution of experimental materials (AD, PTL, SVS); Execution of field/lab experiments and data collection (AD, PTL, PM, VGD, PP); Analysis of data and interpretation (AD, PTL, ANM); Preparation of manuscript (AD, PTL, ANM, SC).

Declaration

The authors declare no conflict of interest.

Acknowledgement

The authors gratefully acknowledge receipt of the rust inoculum and off season screening facility at ICAR-IIWBR, Regional Station, Flowerdale, Shimla.

References

- Anonymous, AICW&BP Crop protection progress report, 2016-17, ICAR-IIWBR, Karnal.
- Gautam A., Sai Prasad S. V. and Jajoo A. 2013. Heritability and correlation of yield and its contributing traits under terminal heat (late sown) situations in durum wheat. Progressive Res., **8**: 203-208.
- Jain S. K., Bhardwaj S. C., Prashar M. and Singh S. B. 2013. Physiologic specialization and new virulences of *Puccinia graminis* f. sp. *tritici* causing black rust of wheat (*Triticum aestivum*) in India during 2005-2009. Indian J. agric. Sci., **83**: 1058-1063.
- Joshi L. M., Srivastava K. D., Singh D. V. and Ramanujam K. 1980. Wheat rust epidemics in India since 1970. Cereal Rust Bull., 8: 17-21.

- Kolmer J. A. 2003. Postulation of leaf rust resistance genes in selected soft red winter wheats. Crop Sci., 43: 1266-1274.
- Letta T., M. Maccaferri A., Badebo K., Ammar A., Ricci J., Crossa J. and Tuberosa R. 2013. Searching for novel sources of field resistance to Ug99 and Ethiopian stem rust races in durum wheat via association mapping. Theor. Appl. Genet., **12**6: 1237-1256.
- McIntosh R. A., Wellings C. R. and Park R. F. 1995. Wheat Rusts: An Atlas of Resistance Genes. CSIRO Publishing, Melbourne, Vic., Australia.
- McIntosh R. A., Yamazaki Y., Dubcovsky J., Rogers J., Morris C., Appels R. and Xia Xi C. 2013. Catalogue of gene symbols for wheat. In: Proc. 12th Int. Wheat Genet. Symp., 8-13 September 2013, Yokohama, Japan.
- Mishra A. N., Gautam S. S., Sitaram Y., Kaushal K., Dubey V. G. and Sai Prasad S. V. 2015. Sources of resistance to Indian pathotypes of *Puccinia graminis* tritici and *P. triticina* in durum wheat. Plant Breed., **134**: 508-513.
- Mishra A. N., Shirsekar G. S., Yadav S. R., Dubey V. G., Kaushal K., Sai Prasad S. V. and Pandey H. N. 2009. Protocols for evaluating resistance to leaf and stem rusts in durum and bread wheats. Indian Phytopath, 62(4): 461-468.
- Nagarajan S. and Joshi L. M. 1975. A historical account of wheat rust epidemics in India and their significance. Cereal Rusts Bull., **3**: 29-33.
- Olivera P. D., Badebo A., Steven S. Xu., Klindworth D. L. and Yue Jin. 2012. Resistance to Race TTKSK of *Puccinia graminis* f. sp. *tritici* in Emmer Wheat. Crop Sci., **52**: 2234-2242.
- Pink D. A. C. 2002. Strategies using genes for non-durable disease resistance. Euphytica, **124**: 227-236.
- Sumikova T. and Hanzalova A. 2010. Multiplex PCR assay to detect rust resistance genes *Lr26* and *Lr37* in wheat. Czech. Genet. Plant Breed., **46**: 85-89.
- Zia UI Qamar, Urmil K. Bansal and Harbans S. Bariana. 2009. Genetics of stem rust resistance in three durum wheat cultivars. Intern. J. Plant Breed., **3**(2): 99-102.