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Inheritance of white rust in newly developed resistant line of Brassica Juncea from local germplasm

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Abstract
Brassicas are important source of vegetable oils in all over the world but various abiotic and biotic stresses caused huge losses to yield out of which white rust occurred 50 to 89.8%. The cultivation of disease resistant varieties seems to be the most practical and affordable means to combat against this disease. In the present study, new white rust resistant line of Indian mustard PWR 15-8 was screened out from local germplasm collected from hills of Uttarakhand (India). The inheritance of white rust in PWR-15-8 was deciphered by crossing it with two agronomically superior susceptible varieties. All F_{1s} were found resistant and F_2 gave segregation of 3:1 for resistant and susceptible plants indicating thereby that the resistance is monotonically dominant over susceptibility. As expected the observations on back crosses of F_1s with their respective susceptible strains (BC_2) showed segregation of 1: 1, while all BC_1 plants were resistant.

Keywords: white rust Indian mustard resistance inheritance

Introduction
Oilseed *Brassicas* are important source of vegetable oils as 15 % contribution to world oilseed production met by them. In India, these crops occupy premier position with a contribution of about 28% of the total oilseeds production. Though, world production rose of this crop but still the demand for rapeseed-mustard oil continues to escalate steeply due to increasing consumption and diversion of bioenergy use. Biotic and abiotic factors are one of the major constraints for low productivity of rapeseed-mustard. These crops are harmed by various diseases like, *Alternaria* blight, white rust, downy mildew and *Sclerotinia* rot at various phases of plant development. White rust was caused by the parasite *Albugo candida* (Pers. Ex Lev.) Kuntze, has as of late turned into the most widespread and ruinous infection in India. All aerial plant parts had demonstrated the manifestations of assault. The disease shows up as conspicuous white pustules on the leaves, stems and inflorescence. Stag heads (inflorescence nerves) may likewise seem later in the developing season because of contamination of meristematic host tissue (Verma and Petrie, 1980) [11]. Noteworthy yield misfortunes because of white rust contamination have been reported (Kumari *et al*. 1970) [3]. This oomycetes pathogen causes 50 to 89.8 per cent misfortunes in seed yield. Most of release cultivars are defenceless to this disease. The chemical control has not been found much effective. Therefore, cultivation of disease resistant varieties seems to be the most practical and affordable means to combat against this disease. Some sources of resistance have already been identified however; the hereditary control for white rust isn't completely comprehended (Thakral and Singh, 1986) [9]. Information on the nature and mode of inheritance of genes controlling resistance and their stability under various agroclimatic conditions is imperative for effective utilization of genetic resistance against diseases in the breeding programme (Chauhan and Sharma, 2001) [2]. In this way, focused research efforts toward this direction made under AICRP on Rapeseed-mustard have led to the identification of several sources for resistance to white rust disease. Despite the fact that several sources of resistance have been described in *Brassica juncea (*Saharan *et. al*, 1988) [6], the information on incorporation of resistance to agronomically superior cultivars has not been reported. Therefore, it is essential to understand the nature of genetic resistance against white rust diseases in available sources. In the present study, an attempt has been made in this direction using a new resistant source namely PWR 15-8 in crosses with RGN 73 and PM-25, which are agronomically superior but susceptible to

white rust disease, and also to formulate suitable breeding strategy for improvement of Indian mustard.

Material and Method

Cultivars

The resistant line to white rust PWR-15-8 used in the present study was selected from mustard germplasm collected from Uttarakhand hills (GP-11-22) from village-Van, Block-Dewal of district Almora (Altitude 7612 m, longitude 79° 37.454; Latitude 30° 11.45). This germplasm was grown in the field of Norman E. Borlaug Crop Research Centre at G. B. Pant University of Agriculture & Technology, Pantnagar, Uttarakhand during 2011-12 where it showed segregation for several traits including reaction against white rust disease, and selection under field conditions yielded 22 plants with resistant to white rust. Further screening from 2011-12 to 2014-15 in the field under artificially inoculated conditions resulted in 14 promising lines free from white rust infection at cotyledonary stage. At true leaf stage, only three lines showed resistant while only PWR-15-8 was found highly resistant (Annual Progress Report, 2015-16) [1] . RGN-73 and PM-25 were the susceptible but agronomically superior line used in this study.

Crossing and advancement of generations

Two crosses namely PWR 15-8×RGN73 and PWR 15-8×PM25 were made using selected parental lines during *rabi* 2014-15. During 2015-16, the seeds of F_1 crosses along with their parents were grown in un-replicated plots. These F_1's were crossed with respective parents (P_1 and P_2) to produce back cross seed. The F_1's were self-pollinated to produce F_2 seeds. At the same time fresh F_1 crosses were also made to obtain sufficient quantity of F_1 seeds for the final experiment. Thus, the family of each cross comprised of six generations viz., P_1, P_2, F_1, F_2, BC_1 and BC_2. The disease incidence of white rust was worked out on the basis of rating scale (0-9) and disease severity. The plants falling under 0 to 5 rating were grouped as resistant plants, while plants with disease ratings >5 fall were grouped under susceptible category.

For assessment of reaction of parents and different generations to white rust diseases, three leaves per selected plant were randomly taken from each generation of six crosses to record the percent leaf area infected using 0-9 point scale being used under AICRP on Rapeseed-Mustard.

Rating	Disease reaction	Leaf area infected (%)
0	Immune	No infection
1	Highly resistant	1-5% leaf area infected
3	Resistant	5-10% leaf area infected
5	Moderately resistant	11-25% leaf area infected
7	Susceptible	26-50% leaf area infected
9	Highly susceptible	>50% leaf area infected

Resistant and Susceptible plant were selected in the field condition following aforementioned criterion. Further verification of resistance was carried out in the lab for which healthy leaves from rosette of 12-14 days old *B. juncea* seedlings were detached and transferred to petri dishes containing 20-25 ml of autoclaved medium consisting of 0.5 ppm benzyl adenine and 0.8% agar. Leaves were placed in the dishes within 15 minutes of detachment. Leaves were drop-inoculated with a zoospore suspension derived from zoosporangia of *A. candida.* Control leaves were treated with distilled water. Leaves were kept under 100% relative humidity for 72-h with day- night temperatures of 21 and 16°C, respectively (Verma and Petrie, 1978) [10]. After 8to 12 days observations were recorded.

Statistical analysis

The chi square method (Snedecor and Cochran, 1967) [7] was used to test the goodness of fit of segregation F_2 and test crosses populations with the excepted phenotypic mono and dihybrid ratios.

Result and Discussion

Reaction of different generations of two crosses for white rust disease has been presented in Table 1. Results showed that all the plants in all the F_1s were resistant and resembled the resistant line used as P1 (PWR 15-8), suggesting that resistance in this line is a dominant trait. The distribution of resistant and susceptible plants in F_2 generation gave a segregation of 3 resistant: 1 susceptible plants, indicating thereby that the resistance is monotonically dominant over susceptibility. As expected the observations on back crosses of F_1s with their respective susceptible strains (BC_2) showed segregation of 1 resistant: 1 susceptible plants, while all BC_1 plants were resistant. These results confirm the single gene control of white rust resistance in PWR 15-8. These results are similar to that of (Paladhi *et. al.,* 1993), (Sridhar and Raut 1998), (Sachan *et. al.,* 2000) and (Chauhan and Sharma, 2001) [4, 8, 5, 2]. However, based on the results of this study it is difficult to ascertain whether genes conditioning resistance to white rust resistance in this line is same or different from those used in earlier studies. Dominant nature of resistance in PWR 15-8 line also suggest that transfer of resistance to agronomically superior strains/lines should be straightforward following backcrossing followed by pedigree method of breeding. To further confirmed above results, we used leaf detached method where 2 weeks old leaves of each generation subjected to white rust inoculum collected from field of *Brassica juncea* L. This procedure was carried out in laboratory condition to ensure the resistant capability of the resistance generation. The same results were obtained i.e. resistant individuals showed resistant against white rust in laboratory conditions as well. Therefore, it verified the single gene control of white rust resistance in PWR-15-8, line of mustard (Fig. 1).

Table 1: Segregation for white rust resistant and susceptible plants in different generations of two crosses in Indian mustard

Cross	Generations	Total plants	Number of plants		Expected Ratio (R: S)	$\chi 2$ Value	Probability
			Resistant (R)	Susceptible (S)			
PWR 15-8 × RGN73	P1	60	All	-	1 : 0	-	-
	P2	60	-	All	0 : 1	-	-
	F1	60	All	-	1 : 0	-	-
	F2	240	173	67	3 : 1	1.09	0.20-0.30
	BC1	150	All	-	1 : 0	-	-
	BC2	150	80	70	1 : 1	0.66	0.30-0.50
	P1	60	All	-	1 : 0	-	-

	P2	60	-	All	0 : 1	-	-
PWR 15-8 × PM25	F1	60	All	-	1 : 0	-	-
	F2	240	175	65	3 : 1	0.56	0.30-0.50
	BC1	150	All	-	1 : 0	-	-
	BC2	150	78	72	1 : 1	0.24	0.50-0.70

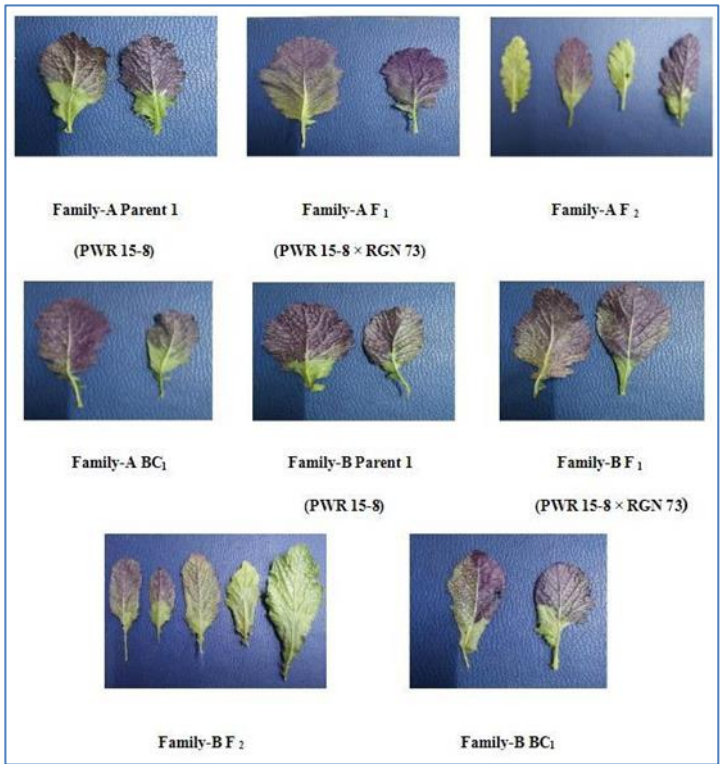

Fig 1: White rust resistance tested under laboratory conditions using inoculum

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