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Assessing the Disease Severity of Alternaria Blight of Rapeseed-Mustard in Jammu Province of J&K and Screening of Germplasm against the Disease

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A B S T R A C T

Alternaria blight of rapeseed-mustard caused by *Alternaria brassicae* (Berk) Sacc. is one of the important diseases of rapeseed-mustard in Jammu division of J&K. Survey conducted revealed that maximum disease severity in leaves at 75 DAS and 100 DAS was 38.53 and 49.71 per cent and in pods was 32.72 and 44.97 per cent, respectively, observed in District Reasi. However, Samba recorded least disease severity at 75 and 100 DAS with 20.40 and 32.97 per cent in leaves and 11.30 and 23.42 per cent in pods. Of the twenty seven different genotypes screened for their reaction against Alternaria blight, two genotypes viz. RH-8113 and PC-5 showed moderate resistance, four genotypes viz. GM-3, RH-1359, RH-819 and JM-1 were found to be moderately susceptible, eighteen genotypes viz. Geeta, PusaBahar, Rohini, RH-30, Shivani, RH-781, RGN-13, GM-2, RRN-505, Krishna, GM-1, PusaJaganath, Vaibhav, RSPN-602, DGS-1, RSPN-25, RSPN-2 and RSPR-69 recorded susceptible reaction. However, three genotypes viz. Kranti, Varuna and CS-54 were found to be highly susceptible.

Introduction

In India, oilseeds constitute the second largest agricultural commodity after cereals and account for nearly 5 per cent of gross national product (GNP) and 10 per cent of the value of all agricultural products (Saharan and Mehta, 2002). Oilseeds are mainly grown for edible oils, spices, condiments and fodder for livestock. In Jammu and Kashmir state, the total area under oilseeds is 63.27 thousand ha, out of which Jammu division has a share of 16.38 thousand ha and the total production of the state is 535q (Anonymous, 2008). Rapeseed-mustard is one of the most important oilseed crops and occupies a prominent place both in terms of area and production. It is a group of crops that contributes 32 per cent of total oilseed production in India and is the second largest indigenous oilseed crop.

Out of 73.09 m tons of estimated rapeseed-mustard produced over 37.00 m ha in the world, India produces 7.90 m tons from 6.70 m ha with 1188 kg/ha productivity (Anonymous, 2014). It is projected that by 2020, 41 per cent (14 million tons) of total demand for oilseed in India will be met by mustard alone (Kalyan *et al.*, 2007).

Despite the fact that India is one of the leading oilseed producing countries of the world, it is not able to meet the edible oil requirement for its vast population. Among different constraints in the production of rapeseed-mustard which is an important oilseed crop, diseases are the most important limiting factors which restrict the cultivation and decrease the productivity of these crops. In Indian context, fungal diseases are rated as one of the most important factor contributing to yield losses in oilseed crops (Anita and Gowthaman, 2003).

More than thirty diseases are known to occur on Brassica crops in India. These include Alternaria blight, white rust, downy mildew, powdery mildew etc. Among these, Alternaria blight caused by *Alternaria brassicae* (Berk.) Sacc. has been reported from all the continents of the world affecting most cruciferous crops and is one among the important diseases of rapeseed-mustard causing severe yield losses with no proven source of transferable resistance in any of the hosts (Meena *et al.,* 2010). The disease occurs regularly year after year during cropping season in severe form and infects both leaves as well as siliquae thereby resulting in reduction of quantity and quality of the crop (Saharan, 1992).

A. brassicae, infects the aerial plant parts causing cholorotic and necrotic foliar lesions (Verma and Saharan, 1994).Besides quantitative loss in yield, the quality of seed i.e., seed size, colour and germination are also drastically affected due to this disease (Randhawa and Aulakh, 1981). Yield losses up to 71.4 per cent (Saharan *et al.,* 2003) and losses in oil content to the tune of 14.6-36 per cent have been reported (Ansari *et al.,* 1988).

Management of Alternaria blight is very difficult and requires frequent fungicidal sprays. Although, crop rotation helps in avoiding the soil-borne primary inoculum but is practically not feasible. Keeping in view, the environmental hazards associated with the use of chemicals, cultivation of varieties resistant to the disease is a better option for its management. Keeping in view, the environmental hazards associated with the use of chemicals, cultivation of varieties resistant to the disease is a better option for its management.

Materials and Methods

Field experiments of the present investigation on Alternaria blight of rapeseed-mustard were conducted at the University Research Farm, Chatha. The laboratory experiments were carried out in the Division of Plant Pathology. The detailed account of materials and methodology adopted is as follows:

Survey and Surveillance

Survey of rapeseed-mustard growing area of Jammu Division was conducted to monitor the prevalence and status of Alternaria blight for which five districts were selected on the basis of their area under the cultivation of the crop. Three villages in each district were identified and five fields of every village were marked for assaying the status of the disease. The disease was recorded using quadrant ($1m^2$) at 4-5 spots in each field. Randomly five plants were selected and tagged for taking observations.

Five leaves and five pods per plant were taken from different plant parts for scoring the disease intensity. Observations for disease severity were taken at 75 and 100 days after sowing. The overall disease scoring was done at 0-6 rating scale on the basis of disease assessment key for Alternaria blight in rapeseed-mustard (Conn *et al.,* 1990) (Fig. 1).

Disease severity was calculated using the following formula:

$$\text{Disease severity (\%)} = \frac{\text{Sum of all disease ratings}}{\text{Total number of ratings} \times \text{Maximum disease rating}} \times 100$$

Isolation, purification and maintenance of pathogen

Different infected plant parts *viz*. leaves, pods and stems of infected rapeseed-mustard plants were collected in paper bags and brought to the laboratory for further investigation.

The bits of diseased portion of infected plant parts along with healthy portion were cut into bits of 8-10 mm, surface sterilized with sodium hypochlorite solution (0.1%) for 30 seconds, washed thrice with sterilized distilled water and thereafter three-four bits were placed in each petriplate containing Potato Dextrose Agar (PDA) medium.

Identification, purification and maintenance of pathogen

The inoculated plates were incubated in BOD incubator at 22 ± 2°C and regularly monitored for the fungal growth. The fungus was identified on the basis of morpho-cultural characteristics (Barnett and Hunter, 1972).

Pure culture of the fungus was obtained by single hyphal tip method (Rather, 2005). For confirmation of the identity of fungus, the slants of pure culture were sent to National Bureau of Agriculturally Important Microbes (NBAIM), Mau. Sub culturing of pure slants of the fungus was done regularly at 15 day intervals.

After every three sub-culturing, the spore suspension of *Alternaria brassicae* was sprayed on live hosts and fresh isolations were made from artificially inoculated diseased plant parts in order to maintain the viability of spores.

Preparation of spore suspension

For inoculation, spore suspension was prepared from freshly developed conidial growth using sterile distilled water and then strained through muslin cloth. The spore concentration was adjusted to $1x10^5$ conidia/ml distilled water using hemocytometer. The plants were sprayed with freshly prepared spore suspension using an atomizer (Vishunavat and Kolte, 2008).

Screening of germplasm

Germplasm for screening of rapeseed-mustard against Alternaria blight was obtained from Directorate of Rapeseed-Mustard Research (DRMR), Bharatpur and was raised in triplicate in rows each of 3m length with a susceptible check Varuna after every two test rows. Experimental field where rapeseed-mustard was grown during previous years was used for the present investigation. To create maximum disease pressure in the field, repeated inoculations at 35, 50 and 65 Days After Sowing (DAS) of spore suspension as discussed in 2.3.1 on the rapeseed-mustard plants were given and higher dose (80 kg/ha) of nitrogen was applied. The crop spacing used was 30 x 10 cm following standard package of practices (Anonymous, 2007). Randomly five plants from each row were selected and tagged for taking observations for initial appearance of disease symptoms and the disease severity at 75 and 100 days after sowing as discussed in 2.1.

Results and Discussion

Survey and surveillance

To assess the disease severity of Alternaria blight of rapeseed-mustard in different districts of Jammu Division, extensive periodic surveys were conducted during Rabi season at different villages of District Jammu,

Kathua, Samba, Reasi and Udhampur. The data presented in Table 1 revealed that the disease was encountered in all the locations surveyed at 75 Days after sowing (DAS) and 100 DAS. The maximum disease severity observed in leaves was 38.53 and 49.71 per cent and in pods was 32.72 and 44.97 per cent at 75 and 100 DAS respectively reported from District Reasi followed by Udhampur (49.54% in leaves and 37.68% in pods), Jammu (40.36% in leaves and 28.73% in pods) and Kathua (34.54% in leaves and 31.24% in pods) districts at 100 DAS. However, Samba recorded minimum disease severity of 20.40 and 32.97 per cent in leaves and 11.30 and 23.42 per cent in pods at 75 and 100 DAS respectively.

Identity of the pathogen

The fungal culture submitted to NBAIM, Mau was identified as *Alternaria brassicae* and the culture has been deposited in Culture Collection Bank of NBAIM vide Accession Number NAIMCC-F-02179.

Screening of germplasm

Trial for screening and evaluation of rapeseed-mustard germplasm under Jammu conditions was laid during Rabi season at University Research Farm, Chatha. Twenty seven different genotypes of rapeseed-mustard germplasm were sown to test their reaction against Alternaria blight. Leaves and pods of the genotypes under test were scored for disease severity at 75 and 100 DAS using the scale proposed by (Conn *et al.*, 1990). Of the germplasm tested, two genotypes *viz.* RH-8113 and PC-5 showed moderate resistance at 75 and 100 DAS (>10-20% disease severity), four genotypes *viz*. GM-3, RH-1359, RH-819 and JM-1 were found to be moderately susceptible (>20-30% disease severity) while eighteen genotypes *viz.* Geeta, PusaBahar, Rohini, RH-30, Shivani, RH-781, RGN-13, GM-2, RRN-505, Krishna, GM-1, PusaJaganath, Vaibhav, RSPN-602, DGS-1, RSPN-25, RSPN-2 and RSPR-69 were found to be susceptible recording a disease severity ranging from 30 to 50 per cent in leaves and pods at 75 and 100 DAS. Three genotypes *viz.* Kranti, Varuna and CS-54 were found to be highly susceptible to Alternaria blight recording a disease severity of more than 50 per cent at both 75 and 100 DAS.

Extensive survey of rapeseed-mustard growing areas of Jammu Division of Jammu and Kashmir was conducted to monitor the prevalence and severity of Alternaria blight. Survey was conducted in fifteen villages of five different districts and observations on disease severity were recorded at 75 and 100 DAS on leaves as well as pods of rapeseed-mustard. Perusal of data in Table 1 showed that the disease was observed in all the locations with varying ranges at both the stages of observation. Maximum disease severity at 100 DAS was recorded in Reasi (49.71% in leaves and 44.97% in pods) followed by Udhampur (49.54% in leaves and 37.68% in pods), Jammu (40.36% in leaves and 28.73% in pods) and Kathua (34.54% in leaves and 31.24% in pods). The least affected district was Samba (32.97% in leaves and 23.42% in pods).

In the present studies, disease severity ranging from 32.42 to 54.94 per cent in leaves and 21.54 to 46.98 per cent in pods was recorded from different districts of Jammu division. Losses due to this disease have been reported from India and world by different workers. Kolte (1985) reported 10-75 per cent losses in yield of oil yielding crops from India. The seed production of Brassicas has been reported to be greatly reduced by the attack of this disease which invades siliquae and penetrate the seeds besides damaging the assimilatory tissues of the leaves and stem (Bains and Tewari, 1986).

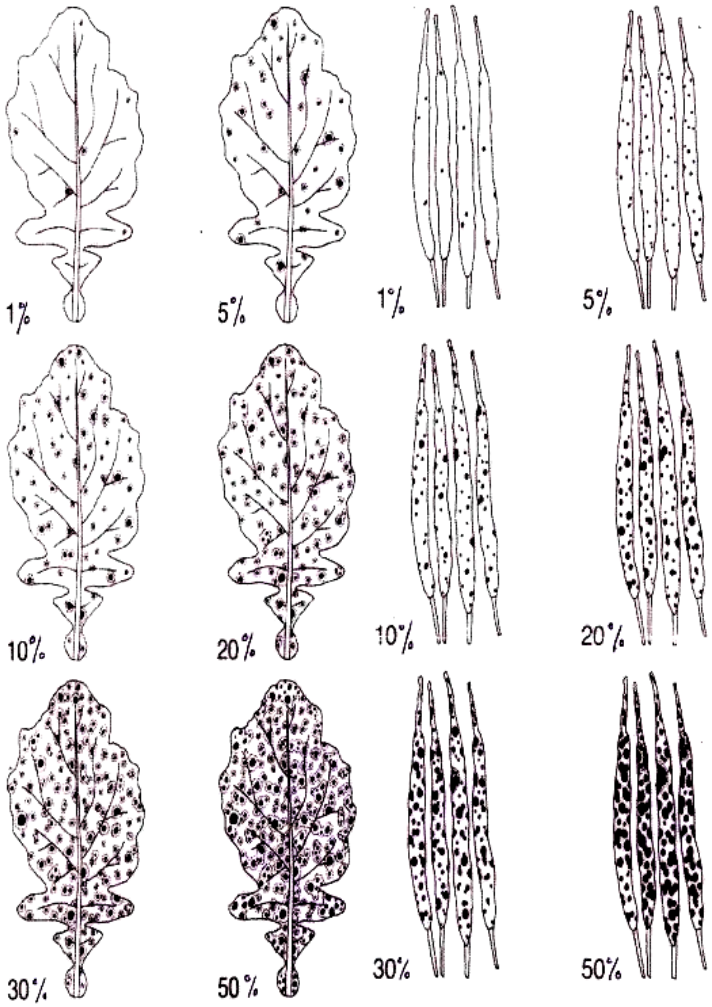
Table.1 Severity of Alternaria blight of rapeseed–mustard in different districts of Jammu Division

Location	Per cent disease severity			
	Leaf		Pod	
	75 DAS	100 DAS	75 DAS	100 DAS
Jammu				
R.S. Pura	23.44	36.28	13.04	22.84
Akhnoor	32.84	41.46	23.62	30.64
Chatha	34.24	43.36	22.72	32.72
Range	23.44-34.24	36.28-43.36	13.04-23.62	22.84-32.72
Mean±S.E.(m)	**30.17±0.544**	**40.36±0.431**	**19.79±0.411**	**28.73±0.977**
Kathua				
Chadwal	23.30	32.42	12.32	21.84
Nagri	20.24	38.68	11.20	35.90
Jarai	20.26	32.54	11.92	36.00
Range	20.24-23.30	32.42-38.68	11.20-12.32	21.84-36.00
Mean ±S.E.(m)	**21.26±0.384**	**34.54±0.764**	**11.81±0.208**	**31.24±0.272**
Samba				
Nud	20.94	34.28	10.08	21.54
Banglarh	21.52	30.80	12.04	22.82
Sumb	18.74	33.84	11.80	25.92
Range	18.74-21.52	30.80-34.28	10.08-12.04	21.54-25.92
Mean ±S.E.(m)	**20.40±0.268**	**32.97±0.656**	**11.30±0.161**	**23.42±0.556**
Reasi				
SirlaBhaga	36.94	43.48	27.60	42.86
Gran Morh	40.62	54.94	31.68	46.98
Seela	38.04	50.72	38.89	45.08
Range	36.94-40.62	43.48-54.94	27.60-38.89	42.86-46.98
Mean ±S.E.(m)	**38.53±0.231**	**49.71±0.777**	**32.72±0.182**	**44.97±0.228**
Udhampur				
Chari Suwail	32.60	49.84	22.02	36.10
SialSallan	34.50	50.94	21.86	39.96
Battalwalian	35.80	47.86	20.20	36.98
Range	32.60-35.80	47.86-50.94	20.02-22.02	36.10-39.96
Mean± S.E.(m)	**34.30±0.453**	**49.54±0.729**	**21.36±0.193**	**37.68±0.235**
Overall range	18.74 –40.62	32.42– 54.94	11.20-38.89	21.54-46.98
Grand mean± S.E.(m)	28.93±0.376	41.42±0.671	19.39±0.231	33.20± 0.454

Table.2 Reaction of rapeseed-mustard germplasm against Alternaria blight under Natural conditions

Germplasm	Initial appearance of disease (DAS)	Per cent disease severity					
		Leaves			Pods		
		75 DAS	100 DAS	Reaction	75 DAS	100 DAS	Reaction
Geeta	45	14.0	39.1	S	11.9	31.8	S
GM-3	50	12.2	28.2	MS	7.5	22.1	MS
RH-1359	45	11.7	23.5	MS	10.7	20.7	MS
Kranti	42	35.7	60.5	HS	23.2	56.3	HS
PusaBahar	50	25.4	38.1	S	24.2	35.5	S
RH-819	48	16.1	28.2	MS	19.5	29.2	MS
Rohini	50	24.9	42.2	S	24.2	37.7	S
RH-30	46	28.7	47.4	S	24.7	37.1	S
Shivani	49	29.5	46.7	S	23.3	39.8	S
RH-781	43	27.1	48.7	S	23.1	37.5	S
Varuna	40	38.4	62.3	HS	35.6	54.2	HS
RGN-13	50	31.7	48.4	S	14.3	38.3	S
GM-2	51	27.2	40.5	S	15.4	37.9	S
CS-54	53	26.9	59.2	HS	18.5	50.3	HS
JM-1	50	12.2	28.8	MS	10.2	22.3	MS
RRN-505	48	20.7	35.3	S	15.3	31.6	S
Krishna	48	28.5	40.3	S	13.9	36.7	S
RH-8113	57	8.5	18.2	MR	7.3	13.1	MR
GM-1	50	27.1	45.5	S	23.2	36.4	S
PusaJaganath	49	28.7	46.2	S	15.1	32.7	S
Vaibhav	51	28.3	39.7	S	18.8	35.2	S
RSPN-602	55	29.0	47.8	S	25.5	37.4	S
PC-5	58	6.9	18.2	MR	5.2	13.5	MR
DGS-1	50	29.2	46.9	S	15.7	33.4	S
RSPN-25	52	27.6	48.7	S	10.8	32.5	S
RSPN-2	50	26.4	43.1	S	19.9	35.5	S
RSPR-69	52	29.8	45.5	S	20.2	34.7	S

Fig.1 Disease assessment key for Alternaria Blight in rapeseed-mustard [12]



Rating	Leaf and Pod	Reaction
0	No infection	**I**
1	Upto 5% area covered	**HR**
2	>5-10 % area covered	**R**
3	>10-20 % area covered	**MR**
4	>20-30% area covered	**MS**
5	>30-50 % area covered	**S**
6	**>50 % area covered**	**HS**

Where, I = Immune, HR = Highly resistant, R = Resistant, MR = Moderately resistant, MS = Moderately susceptible, S = Susceptible and HS = Highly susceptible

Plate 1. Symptoms of Alternaria blight on mustard leaves

Plate 2. Symptoms of Alternaria blight on of mustard on pods

Plate 3.Field view of screening of germplasm of rapeseed-mustard.

Kadian and Saharan (1983) and Ram and Chauhan (1998) reported 15 to 71 per cent losses in rapeseed-mustard due to Alternaria blight. The disease is also reported to be of major economic importance in Australia, France, Poland and of moderate importance in Canada and United Kingdom (Barman and Bhagwati (1995). Hong and Fitt (1996) reported that Alternaria blight results into yield losses up to 71.4 per cent. Bharti *et al.*, (2016) also observed the highest disease intensity of 53.60 per cent from Kalyanpur (Kanpur) and lowest intensity (37.60 %) was recorded from Bagha (Kanpur) during their survey of various locations. During extensive surveys, Gupta *et al.*, (2017) found the maximum disease intensity (44.13 %) at Chatha while minimum disease intensity (28.90 %) was recorded from Gudwal area of Jammu Province.

For the successful establishment of Alternaria blight of rapeseed-mustard, the causal organism (*Alternaria brassicae*) requires low temperature, high humidity and splashing rain (Humpherson and Phelps, 1989). In India, maximum temperature of 27-28^{0}C, minimum temperature of 14-15^{0}C, average relative humidity more than 65 per cent, intermediate winter rains and wind velocity 2-5 km/hr has

been reported to be most conducive to Alternaria blight development in rapeseed-mustard (Sangeetha and Siddaramaiah, 2007; Conn *et al.,* 1990). Prevailing low temperature coupled with high humidity in Reasi and Udhampur may be the reason for increased severity of the disease in these districts. The pathogen is greatly influenced by weather with the highest disease incidence reported in wet seasons and in areas with relatively high rainfall (Meena *et al.,* 2010). Alternaria blight of rapeseed-mustard has been reported to be predominantly seed borne (Parajuli, 2005). Thus, it seems that the disease problem has aggravated due to continuous use of infected local seed that has not been replaced for a number of years and no plant protection measures adopted at appropriate stages along with improper cropping practices being followed by farmers.

Twenty seven genotypes were procured from DRMR, Bharatpur and evaluated for their reaction towards Alternaria blight so as to screen out the germplasm resistant to Alternaria blight for this region. Observations for leaf and pod blight were taken at 75 and 100 DAS. During the experiment, it was found that out of all the genotypes screened, two genotypes *viz.* RH-8113 and PC-5 showed moderate resistance against the disease (>10-20%) while four genotypes *viz*. GM-3, RH-1359, RH-819 and JM-1 showed moderate susceptibility (>20-30%) and eighteen genotypes (Geeta, PusaBahar, Rohini, RH-30, Shivani, RH-781, RGN-13, GM-2, RRN-505, Krishna, GM-1, PusaJaganath, Vaibhav, RSPN-602, DGS-1, RSPN-25, RSPN-2 and RSPR-69) were found to be susceptible recording disease severity ranging from 30-50 per cent. Three genotypes *viz.* Kranti, Varuna and CS-54 showed high susceptibility and the disease severity was found to be more than 50 per cent (Table 2). Sources of resistance to the disease in *Brassica napus* and *Brassica juncea* have been listed by (Saharan *et al.,* 1988) and the cultivar Prakash has been reported to be highly susceptible. It has been reported that genotypes *viz.* DIR-1507 and DIR-1522 of *Brassica juncea* had stable resistance against Alternaria blight (Dang *et al.,* 2000). Fifty four lines/varieties in Alternaria sick plot were tested by Srivastava *et al.,* (2001) and observed that none of the varieties was resistant to Alternaria blight. Among the various varieties tested by Mondal (2008) Jhumkta, Sanjuka, Aseech, Seeta and Bhagirathi were found to be better and could be recommended against the disease. The cultivars Binoy, Agrani, Panchali and SwamaSarisa showed more susceptible reaction against Alternaria blight than rest of the cultivars. Screening of sunflower genotypes to evaluate them for resistance to *Alternaria helianthi* was performed and it was found that the disease intensity for hybrids ranged from 3.73 to 52.33 per cent. RHA 587 and ARG x RHA 587 were found to be resistant to Alternaria blight both under field and laboratory conditions and therefore have the potential to reduce yield losses because of this disease in the field (Reddy *et al.,* 2006). It can be concluded that use of germplasm which showed resistance to Alternaria blight during the present studies, can check the disease severity as well as the disease spread under Jammu conditions and thereby minimizing the losses incurred by farmers due to Alternaria blight of rapeseed-mustard.

Present studies were undertaken on Alternaria blight of rapeseed-mustard in order to determine the status of disease in Jammu Division, to screen germplasm of rapeseed-mustard for resistance to Alternaria blight.

During the course of survey, the disease was found to be present at 75 and 100 DAS in all the districts. Maximum disease severity reported was 38.53 per cent in leaves and 32.72 per cent in pods at 75 DAS and 49.71

per cent in leaves and 44.97 per cent in pods at 100 DAS which was observed in Reasi district. However, minimum disease severity i.e. 20.40 per cent and 11.30 per cent in leaves and pods respectively at 75 DAS and 32.97 per cent and 23.42 per cent in leaves and pods respectively at 100 DAS was observed in Samba.

The results of screening experiments of rapeseed-mustard germplasm revealed that out of twenty seven genotypes tested for disease reaction against Alternaria blight, two genotypes *viz.* RH-8113 and PC-5 were found to be moderately resistant at 75 and 100 DAS (>10-20% disease severity), four genotypes *viz*. GM-3, RH-1359, RH-819 and JM-1 were found to be moderately susceptible (>20-30% disease severity), eighteen genotypes *viz.* Geeta, PusaBahar, Rohini, RH-30, Shivani, RH-781, RGN-13, GM-2, RRN-505, Krishna, GM-1, PusaJaganath, Vaibhav, RSPN-602, DGS-1, RSPN-25, RSPN-2 and RSPR-69 were found to be susceptible recording a disease severity ranging from 30 to 50 per cent in leaves and pods. However, three genotypes *viz.* Kranti, Varuna and CS-54 were found to be highly susceptible to Alternaria blight (>50% disease severity) at 75 and 100 DAS.

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