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LOCATION ASSESSMENT OF *FUSARIUM OXYSPORUM F. SP. CICERI* IN SEEDS OF DIFFERENT CULTIVERS OF CHICKPEA

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ABSTRACT In present studies six varieties (3 susceptible and 3 resistant) namely ICP-1454, ICC-1876, Radhey (susceptible), ICC-203, Avarodhi, and ICCV-32 (resistant to disease under field condition) were taken. The pathogen was detected in the all-susceptible varieties of chickpea ranging from 12-20 present with maximum in ICP-1454(20%) followed by Radhey (19%) and ICC-1876 (17%). It was not present in resistant variety except Avarodhi. (3%) Varieties ICC-203 and ICCV-32 did not carry seed infection.

KEYWORDS : Chickpea, *Fusarium oxysporum* f. sp. *ciceri*, Varieties, Wilt.

INTRODUCTION

Pulse crops play an important role in Indian agriculture, besides being rich in protein. Sprouted seeds are recommended for breakfast. Its green leaves produces malice and oxalic acids, which are prescribed for intestinal disorders. The availability of pulses per capita is 27 gm./day. Occurrence of chickpea wilt was first described in India by (Butler, 1918). *Fusarium oxysporum* is ubiquitous, asexual species complex. Isolates of Fusarium oxysporum can cause vascular wilt or cortical rot in many agricultural crops and have been classified into forma specials based on their host specificity (Nelson *et al.,* 1981). Erwin (1958) reported that the foliage of *F. oxysporum* f.sp. *ciceri* infected plant turns yellow before wilting and the xylem tissue shows light brown discoloration.

They sustain the productivity of the cropping system. Their ability to use atmospheric nitrogen through biological nitrogen fixation (BNF) is economically sounder and environmentally acceptable. Pulses are grown 60 per cent in *Rabi* and 40 per cent in K*harif*. Gram (Chana) (*Cicer arietinum* L.) belongs to the sub family Papilionaceae of the family Leguminosae is a herbaceous annual, having branching close to the ground with semi-erect to semi-spreading habit. Gram possesses a highly economic nutritive value as is evident from its analysis. One hundred-gram chickpea seeds contain about 9.8 per cent moisture, 21.1 g protein, and 5.3 g fat. 61.0 g carbohydrates and 3.9 g fiber in addition to vitamins and other things. Taking into consideration the rapid growth of population and alarming situation of protein deficiency in the country, there is need of coordinated efforts by the breeders, agronomists and plant pathologists for making improvement of this important pulse crop.

This crop (chickpea) is subjected to attack by a number of fungal, viral, bacterial and nematode diseases. Inadequate information is available on the *Fusarium* wilt of gram as regards to seed-borne nature. Out of these maladies, wilt alone causes considerable loss to gram in India every year. It is a complex problem and various pathogens as well as physiological factors are implicated to cause wilt of gram, which lead to variation in disease syndrome, although often there is overlapping of symptoms. However, the typical vascular wilt caused by *F. oxysporum* f. sp. *ciceri* is the outstanding problem in Uttar Pradesh.

Haware *et al.,* (1982) showed the fungus to be in the hilum of the seed in the form of chlamydospores like structures. Infected seeds play an important role in the long-distance dispersal of the pathogen and in its introduction into *F. oxysporum* f. sp. *ciceri* free soils and geographic areas (Pande *et al.,* 2007).

MATERIAL AND METHODS

Diseased gram plants showing characteristic wilt symptoms were collected from E.B. Legume Research Farm, C.S. Azad University of Agriculture & Technology, Kanpur in the last week of November, 2002 and March, 2003 and brought to the laboratory for the examined of the seed infection from *F. oxysporum* f. sp. *ciceri.* Colony was critically observed on seed *in situ* by using Standard blotter method (ISTA, 1999). These observations were also recorded by Agar plate method (Neergaard, 1979) for the confirmation of results obtained by blotter method. For both methods (SBM and APM) six varieties (3 susceptible and 3 resistant) namely ICP-1454, ICC-1876, Radhey (susceptible), ICC-203, Avarodhi, and ICCV-32 (resistant to disease under field condition) were taken.

A. Standard blotter method (SBM):

Three hundred seeds of each variety was randomly select from properly homogenize samples, each sample was mixed and divide by precision divider (Gamet type) and then each sample further divided into three lots, each lot contain one hundred seeds. First lot of one hundred seed remains as untreated (control). Second lot was pre-treat with 1% sodium hypochlorite (NaOCl) for 10 minutes and third lot pre-treat with 0.1 % mercuric chloride ($HgCl_2$) solution for 1 minute. Ten replications were use from each lot (Untreated & Pretreated with NaOCl and $HgCl_2$) of each variety. Each replication was contained 10 seeds. Selected 10 seeds were arranged as 9 seeds in the outer ring and 1 seed in the center place on three pre-moist (with sterile distill water) layers of sterilize blotter paper; kept in 90 mm diameter polypropylene/glass petri-plates. These plates was incubated at 25±1^{0}C under alternating cycle of 12 hours NUV (Near Ultra Violet) light and 12 hour darkness for seven days. On eighth day, the seed was examined under stereo-binocular microscope (SBM) at the range on (6.4 x 40 X) for the presence of *F. oxysporum* f. sp. *ciceri*. The numbers of seed bearing the colonies of the test pathogen was count, record and interpret as per cent infection of the test pathogen.

B. Agar plate method (APM):

In this method, Potato dextrose agar medium was is use in the place of blotter paper. One hundred fifty seeds were randomly select from properly homogenize samples, each sample was (variety) further divided, with help of precision divider (Gamet type) into three lots, each lots contain 50 seeds. First lot of 50 seeds was remaining as untreated (control) and remains two lots (50 seeds/each) were treated as given the method in standers blotter method (4.1.2.1). Ten replications was use from each lots (Untreated & Pretreated with NaOCl and $HgCl_2$) of each variety. Each replication was contained 5 seeds. Selected 5 seeds were arranged as 4 seeds in the outer ring and 1 seed in the center place on 90 mm PDA petri plate. Then the plates were incubated for 7 days and next day, the growth of the fungus on seed was examined by necked eye and for specific identification of the test pathogen, use stereo-binocular and compound microscopes. The number of seeds bearing colonies of *F. oxysporum* f. sp. *ciceri* were counts, records and interpret in per cent infection of the test pathogen.

RESULTS AND DISCUSSION

Out of six seed samples, collated from susceptible & resistant cultivars tested for the presence of the pathogen by using standard blotter & agar plate methods, the pathogen was detected in the all susceptible varieties of chickpea ranging from 12-20 per cent. Standard blotter methods had an edge over agar plate method so far as the detection of the pathogen was concerned.

The pretreatment of the seeds with the sodium hypochlorite and mercuric chloride reduced the recovery of the pathogen from the infected seeds. The average recovery percentage from sodium hypochlorite and mercuric chloride pre-treated seeds were 12.17 and 4.67 per cent, respectively. The seed infection was maximum in the susceptible variety ICP-1454 (20%) and minimum resistant variety Avarodhi (3%) whereas two resistant varieties ICC-203 and ICCV-32 did not carry the pathogen. The above finding is similar with the findings of Haware *et al.* (1978), Khune and Patil (1992) and Gangwar (2004).

CONCLUSION

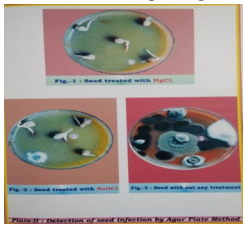
In order to ascertain the extent of *F. oxysporum* f. sp. *ciceri* infection in seeds, seeds of six, varieties *viz.,* ICP-1454, ICC-1876, Radhey, ICC-203, Avarodhi and ICCV- 32 were tested by Standard blotter method and Agar plate method as described under Materials and Methods. The data on per cent seeds infection of *F. oxysporum* f. sp. *ciceri* in different chickpea cultivars in standard blotter method and agar plate methods are presented in **Table-1**.

Table-1: Per cent seed infection of *F. oxysporum* f. sp. *ciceri* in chickpea cultivars in Standard blotter method and Agar plate method.

S. No.	Sample	Standard blotter method			Agar plate method		
		Untreated	Protected with		Untreated	Protected with	
			NaOCl	HgCl2		NaOCl	HgCl2
Susceptible cultivars							
1.	ICP-1454	20	15	6	20	14	5
2.	Radhey	19	13	5	18	12	5
3.	ICC-1876	17	12	3	12	7	4
Resistant cultivar							
4.	Avarodhi	2	1	0	3	0	0
5.	ICC-203	0	0	0	0	0	0
6.	ICCV-32	0	0	0	0	0	0

The results in **Table-1** show that the pathogen *(F. oxysporum* f. sp. *ciceri*) was present in seeds (12-20%) of the varieties of chickpea, which are happened to be susceptible to thus field. The seeds of resistant variety Avarodhi also carried the pathogen to the extent of 3 per cent.

The seeds of three resistant varieties accept Avarodhi did not carry the pathogen. Standard blotter method had an edge over agar plate method **(Plate-II)** so for as the detection of the pathogen was concerned.



The pre-treatment of the seed with the sodium hypochlorite for 10 minutes and 0.1 per cent mercuric chloride for 1 minute reduced the recovery of the pathogen from the infected seeds. The average recovery percentage from sodium hypochlorite and mercuric chloride pre-treated seeds were 12.17 and 4.67 per cent, respective. These were some deference's in the recovery of seed infection in the varieties. It was maximum in the variety ICP-1454 and minimum in the resistant variety Avarodhi whereas two resistant varieties ICC-203, and ICCV-32 did not carry the pathogen.

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