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Studies on the Association of *Fusarium oxysporum* f. sp. *ciceri* with Seeds of Chickpea

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A B S T R A C T

Chickpea wilt caused by *F. oxysporum* f. sp. *ciceri* is one of the most important seed and soil-borne disease in India. The study on association of the pathogen with chickpea seeds showed that the viable mycelium fragments, micro and macro-conidia of the pathogen were present on the seed surface of all susceptible and two resistant varieties/ germplasms in seed washing test. The pathogen was recovered from the untreated and sodium hypochlorite treated seeds of all susceptible varieties/ germplasms in the range of 10.67–18.67 and 5.33–11.33 per cent, respectively in standard blotter method and 8.67–17.00 and 5.00–10.67 per cent, respectively in agar plate method, whereas the seeds of resistant varieties RSG-895 and Phule G-5 also carried the pathogen in untreated and sodium hypochlorite treated seeds. The recovery of pathogen was ranged from 10.0–20.0 per cent from the seed coat, 2.0–8.0 per cent from cotyledons and 2.0–6.0 per cent from embryonal axis of the susceptible varieties/germplasms. Whereas resistant variety RSG-895 carried the pathogen to an extent of 4.0 per cent from seed coat only. The pre-emergence infection of the pathogen was recorded by 6.0–14.0 per cent in different susceptible varieties/ germplasms and 4.0 per cent in resistant varieties during seedling symptom test. The pathogen was survived for 10–15 months in susceptible and 3–5 months in resistant varieties/ germplasms seeds. The pre and post-emergence losses due to the pathogen were ranged from 7.0–12.0 per cent and 6.0–10.0 per cent, respectively in susceptible varieties/ germplasms under field conditions in poly bags. The typical wilt symptoms have developed after 18–22 days of sowing and the average seed infection and seed transmission ratio was 6.08:1 under field conditions in susceptible varieties/ germplasms.

Introduction

Chickpea (*Cicer arietinum* L) is known as Gram or Bengal gram or Spanish pea considered to be the third most important pulse crop of the world. It is cultivated from ancient time in different parts of the world. In India it is an important source of protein in human diet and still the demand of pulses is very high because the majority of people are vegetarian. Chickpea plays a significant role in sustaining the production of subsistence farming system. It is grown on 8.35 million hectares with the production 7.17 million tonnes and productivity 859 kg/ha during 2015-16 in India (Anonymous, 2016). The

total pulses availability in India is 43.8 gm/day/capita. Major production of chickpea comes from central and northern India. However its area and production is also increasing in southern states. Madhya Pradesh stands first as far as acreage and production were concerned in India followed by Rajasthan, Maharashtra, Uttar Pradesh, Karnataka, Andhra Pradesh, Bihar, Haryana, Gujarat, Orissa, West Bengal and Punjab.

The crop is grown in Rabi season and more than 50 pathogens have been reported on this crop from different parts of the world (Nene *et al*., 1996). While the fungal diseases like Fusarium wilt, Dry root rot, Ascochyta blight, Botrytis grey mould and Black root rot are causing maximum damage to the crop in India. Among these diseases of chickpea wilt caused by *Fusarium oxysporum* Schlecht emend. Snyd. & Hans. f. sp. *ciceri* (Padwick) Snyd. & Hans. is a serious seed and soil-borne disease in India (Nene *et al*., 1996; Pande *et al*., 2007). The disease has been prevalent in all the chickpea growing states of the country and due to its regular occurrence the disease influencing successful cultivation of the crop every year. The disease can appear at any growth stages of the plants beginning from seedling to pod stage. It cause on an average 10 per cent loss in yield and the damage has been observed to extent up to 61 per cent and 43 per cent at seedling and adult stages, respectively (Nema and Khare, 1973; Singh *et al*., 1989). The chickpea seeds harvested from wilted plants, when mixed with healthy seeds can carry the wilt pathogen to a new area and can establish the disease in the soil up to economic threshold level with in a three seasons (Pande *et al*., 2007). The mycelium of the pathogen is present in the vascular tissues of the wilt-infected plants (Nema and Khare, 1973). The pathogen was present in the seed coat, cotyledons and embryonal axis and survives upto the extent of 13 months in susceptible cultivars (Gangwar *et al*., 2013). It forms chlamydospores like structures near the hilum region of the seed. Seeds from the wilted plants are generally smaller, wrinkled and discoloured. The pathogen reaches inside the seed by systemic pathways (Haware *et al*., 1978; Conci *et al*., 1985). Singh V.K. (2014) recovered different seed-borne fungi associated with the seed of chickpea by using seed washing test, agar plate method and standard blotter method. The pathogen was recovered from the susceptible plants within a few weeks, the hyphae were inter-and intra-cellular in the pith, xylem and cortex (Kunwar *et al*., 1989). It was isolated from the taproot, lateral root, collar region, main stem, lateral branches and seeds of infected chickpea plants, but not from pod and leaves (Khune and Patil, 1992; Singh and Gangwar, 2017). The pathogen was systemic in nature and can be isolated from all the parts of an infected plant including seed (Nene *et al*., 1979; Harware *et al*., 1996; Gangwar *et al*., 2013; Singh and Gangwar, 2017). Gangwar *et al*., (2013) also reported that the average ratio between seed infection and seed transmission was 6.60:1 amounting 15 per cent of the seed transmission. Therefore, in view of the seriousness of the disease and importance of the crop, the research work was carried out on the association of *Fusarium oxysporum* f. sp. *ciceri* with seeds of chickpea.

Materials and Methods

The study on above mentioned objective was carried out by seed washing test, standard blotter paper method, agar plate method, component plating method, seedling symptom test, serial isolation test and poly bag experiment in the year of 2015 and 2016 at Plant Health Diagnostic Centre, Krishi Vigyan Kendra, Chomu, Jaipur (Rajasthan). A set of twelve chickpea varieties/ germplasm, in which six resistant and six susceptible was selected for these tests. The resistant varieties/germplasms were RSG-895, RSG-

963, RSG-991, Phule G-5, JG-315 and BG-373 and susceptible were ICC-1375, ICC-1876, ICP 1454, Radhey, JG-62 and DGM-1460. The seeds of all susceptible varieties were collected from partially wilted plants.

The seed washing test was used to detect the mycelial fragments and spores (Micro and Macro-conidia) of *F. oxysporum* f. sp. *ciceri* presents on the surface of chickpea seeds. Fifty randomly selected seeds from each variety/ germplasm of both categories were divided into two groups. Each was suspended in 10 ml of sterilized distilled water in the conical flasks, separately. The flasks were shaken by rotary shaker for 10 minutes. The supernatant liquid was decant off from each flask and then the sediment from respective flask was thoroughly mixed in 2.0 ml lactophenol and examined under compound and trinocular zoom microscope for the presence of mycelial fragments and spores (micro and macro-conidia) of *F. oxysporum* f. sp. *ciceri.* Subsequently, their viability tests were also conducted by inoculating the seed washing the PDA medium Petri-plates. The plates were then incubated at 25^0C in a BOD incubator for 7 days. The data of fungal growth were recorded as viability of spores on the surface of the seed.

Standard blotter method (ISTA, 1999) and agar plate method (Neergaard, 1979) were used *in situ* to study the seed infection and growth habit pattern of *F. oxysporum* f. sp. *ciceri* colonies on seed. The above selected 6 resistant and 6 susceptible varieties/ germplasm were taken for both methods. For standard blotter method the seeds of each variety were randomly selected from properly homogenized samples and were divided into two lots; each lot contains one hundred fifty seeds. First lot of one hundred and fifty seeds remained untreated and second was pretreated with 1 per cent sodium hypochlorite (NaOCl) solution for 10 minutes. Fifteen replications were used for each variety, each of ten seeds. These ten randomly selected seeds of each treatment were placed in the sequence of 9+1 at equal distance with the help of sterilized forceps on three layers of moist blotter paper in each 90 mm polypropylene Petri-plates and incubated at 25 ± 1^0C under alternating cycle of 12 hours light and 12 hours darkness for seven days. The seeds were examined after seven days under trinocular zoom microscope for the presence of *F. oxysporum* f. sp. *ciceri*. The number of seeds bearing the colonies of *F. oxysporum* f. sp. *ciceri* were counted, recorded and interpreted as per cent seed infection.

In the agar plate method, potato dextrose agar medium was used in place of blotter papers. One capsule of chloramphenicol 500 mg was added in each flask containing 200 ml of PDA medium before pouring in Petri-plates in order to check the bacterial contamination. One hundred fifty seeds were taken from each variety/ germplasm and randomly divided into two lots (each of 75 seeds): untreated and pretreated with 1 per cent sodium hypochlorite (NaOCl) for 10 minutes. Five randomly selected seeds of each variety/ germplasm from every treatment were placed at equal distance in the sequence of 4+1 at with the help of sterilized forceps on 90 mm PDA Petri-plates under the aseptic condition of bio safety cabinet. A total number of 15 replications for each treatment were taken off. The plates were then incubated at 25 ± 1^0C under alternating cycle of 12 hours light and 12 hours darkness for seven days. The growth of the fungus on seeds was examined after seven days of incubation under trinocular zoom and compound microscopes for specific identification of the pathogen. The number of seeds bearing the colonies of *F. oxysporum* f. sp. *ciceri* were counted, recorded and interpreted in per cent seed infection. The component plating method (Maden *et al*., 1975) was used to study the presence of *F.*

oxysporum f. sp. *ciceri* in different seed components. Seeds of above selected all susceptible varieties/ germplasm and two resistant varieties (RSG 895 and Phule G-5) have taken for the study. These two resistant varieties have been selected on the basis of recovery of the pathogen in standard blotter and agar plate method. The location of *F. oxysporum* f. sp. *ciceri* in seed components have studied on potato dextrose agar medium. Fifty seeds of each variety were soaked separately in the conical flasks containing sterilized distilled water for 24 hours. The seeds were later dissected individually in their components *viz*., seed coat, cotyledons and embryonal axis with the help of sterile needle and forceps in the bio safety cabinet. These seed components were shifted in the Petri-plates containing potato dextrose agar medium under the aseptic condition and incubated at 25 ± 1^0C under alternating cycle of 12 hours light and 12 hours darkness for seven days. The components showing growth of the pathogen was recorded and interpreted in term of per cent infection in seed components.

The pathogen is seed-borne in nature therefore duration of survival in the seed was studied. The seeds were collected from wilted plant at maturity from all susceptible varieties/ germplasms (ICC-1375, ICC-1876, ICP 1454, Radhey, JG-62 and DGM-1460) and two resistant varieties (RSG-895 and Phule G-5). These seeds were stored in paper bags under ambient condition (25 ± 1^0C) in the laboratory in the month of April, 2015. The monthly isolations were taken from the seeds by using agar plate method to know the viability of the pathogen in seed. The data were recorded in the form of survival months of the pathogen in seeds.

Transmission of the pathogen from seed to seedling was studied by using seedling symptom test (Khare *et al.*, 1977), followed by serial isolation method under laboratory conditions (Neergaard, 1979). The infected seeds of all susceptible varieties/ germplasms and two resistant varieties RSG-895 and Phule G-5, collected from partially wilted plants which were taken for seedling symptom test. The one hundred seeds of each variety/ germplasm were surface sterilized with 1 per cent sodium hypochlorite solution for 10 minutes and each seed placed aseptically in separate culture tubes containing 2 per cent water agar medium. These tubes were plugged loosely with cotton and placed in an incubation room at 25 ± 1^0C under 12 hours light and 12 hours darkness for 45 days. Observations on the symptoms on seed and seedling were recorded in the tube after 7 days and up to 45 days and calculated in terms of the ratio of seed infection and seed transmission. Isolations from the diseased tissues and rotted seeds were taken serially on PDA by serial isolation test for confirming the association of pathogen and movement of the pathogen from seed to seedling. Two mm segments were cut from the root to apical tip of the stem and placed on potato dextrose agar medium in Petri-plates and incubated for 7 days at 25 ± 1^0C for confirming the presence of the pathogen.

The poly bag experiment was conducted to study the pre and post emergence losses, number of days required to develop the disease symptoms and the ratio of seed infection and seed transmission. The seeds collected from partially wilted plants of all susceptible and two resistant varieties/ germplasms and were sown directly in 75x45 cm poly bags containing sterilized soil. Five seeds were sown in each bag and transferred in to the shade net house in the month of November, 2015. A total number of 20 bags per variety/germplasm were maintained. The bags were covered with separate polythene to protect them from aerial infection up to 30 days. Thereafter, the covers were removed and begs were shifted into the open area. The need

based irrigation have given with the sterilized water to the bags and seedling emerged were observed daily for the development of the wilt symptoms. The data of per cent seed infection were recorded for untreated seed in the standard blotter method have used in this experiment. The observations on pre and post emergence losses, number of seedlings showing disease symptoms, number of days required to develop typical wilt symptoms were recorded. The seed infection and seed transmission ratio was calculated by using the data of per cent seed infection and number of seedlings showing disease symptoms.

For statistical analysis mean value, standard deviation (SD) and standard error of mean differences (± SEm) was calculated.

Results and Discussion

The results of seed washing test presented in Table 1 indicated that the viable mycelial fragments, micro and macro-conidia of *F. oxysporum* f. sp. *ciceri* were presented on the surface of the seeds of all susceptible varieties/ germplasms *viz*., ICC-1375, ICC-1876, ICP 1454, Radhey, JG-62 and DGM-1460. Where as they were recovered from the seed washing of only two resistant varieties RSG-895 and Phule G-5. The similar results have been reported by earlier workers (Gangwar *et al*, 2013; Haware *et al*., 1978; Khune and Patil, 1992).

The results of standard blotter paper method presented in Table 1 showed that the pathogen was present in untreated seeds of all susceptible varieties/ germplasms in the range of 10.67–18.67 per cent, where as the seeds of two resistant varieties (RSG-895 and Phule G-5) carried the pathogen up to the extent of 6.00–8.33 per cent. The pretreatment of the seeds with 1 per cent sodium hypochlorite (NaOCl) for ten minutes reduced the recovery of the pathogen from the seeds, where it was recovered by 5.33–11.33 per cent from the seeds of all susceptible chickpea varieties/ germplasms and 4.67 and 3.33 per cent from the resistant varieties RSG-895 and Phule G-5, respectively. This indicates that the pathogen was located inside the internal tissues of the infected seeds. The maximum seed infection (11.33%) was recorded in variety JG-62 and minimum (3.33%) in resistant variety Phule G-5. The study also showed that the seed of remaining four resistant varieties were carried the pathogen.

The results of agar plate method presented in Table 1 were also found more or less in same pattern as in the case of standard blotter method. Where the pathogen was recovered from the untreated seeds of all susceptible varieties/ germplasms in the range of 8.67–17.00 per cent. While the pathogen was recovered by 5.67 and 7.00 per cent from the untreated seeds of two resistant varieties Phule G-5 and RSG-895, respectively. The pathogen was recovered in the range of 5.00–10.67 per cent from sodium hypochlorite treated seeds of all susceptible varieties/ germplasms and in the range of 3.00–4.00 per cent from the seeds of resistant varieties Phule G-5 and RSG-895. Whereas results of standard blotter method and agar plate method showed that the seeds of other resistant varieties *viz*., RSG-963, RSG-991, JG-315 and BG-373 did not carry the pathogen. The earlier workers have also reported the seed-borne nature of the pathogen (Conci *et al*., 1985; Gangwar *et al*, 2013; Haware *et al*., 1978; Singh and Gangwar, 2017).

The data presented in Table 2 reveals that the pathogen was present in all seed components of the susceptible varieties/ germplasms. The recovery of pathogen was ranged from 10.0–20.0 per cent from the seed coat of the susceptible varieties. Whereas out of two resistant varieties only one RSG-895 carried the pathogen with the extent of 4.0 per cent

only in seed coat. The pathogen was recovered 2.0–8.0 per cent from the cotyledon of all susceptible varieties/ germplasms. The embryonal axis of different susceptible varieties/ germplasm except DGM-1460 carried the pathogen from 2.0–6.0 per cent. The establishment of infection in cotyledons and embryonal axis was maximum in JG-62. Though the seeds of the resistant variety RSG-895 were not found to carry the pathogen in cotyledons and embryonal axis. The study indicated that the pathogen was carried with the chickpea seeds externally on its surface in the form of mycelial fragments, micro and macro-conidia and internally in the tissues of seed coat, cotyledons and embryonal axis. These results are similar to the findings of earlier workers (Conci *et al*., 1985; Gangwar *et al*., 2013; Haware *et al.*, 1978; Singh and Gangwar, 2017).

The results of seedling symptom test carried out in water agar medium tubes (Table 2) showed that the recovery of *F. oxysporum* f. sp. *ciceri* as a pre-emergence seed infection was ranged from 6.0–14.0 per cent in different susceptible varieties/ germplasms. Whereas it was recovered by 4.0 per cent in the resistant varieties RSG-895 and Phule G-5. The seedlings grown in culture tubes showed the browning of the main roots after 12 days of sowing. Later on after 3 days the secondary roots were also started browning. There after the whole seedling became yellow resulting such seedlings were collapsed and dried. For confirming the presence of pathogen in affected seedlings, the roots and stem pieces were placed on PDA Petri plates and incubated for 7 days. The growth of fungus was observed on eighth day and it was identified as *F. oxysporum* f.sp. *ciceri*. The typical wilt symptoms were observed in 2.0–6.0 per cent of seedlings for different susceptible varieties/ germplasms, while the seedlings of resistant varieties were not showed disease symptoms. The ratio between seed infection and seed transmission was varied from 2.00:1 to 5.00:1 in different susceptible varieties/ germplasms. The maximum initial per cent of seed infection and development of typical wilt symptom was recorded in JG-62. While the ratio between seed infection and seed transmission was highest in ICP-1454. These findings have close concern with the findings of earlier workers (Conci *et al*., 1985; Gangwar *et al*., 2013; Haware *et al.*, 1996; Nene *et al*., 1979).

The results presented in Table 2 on survival of the *F. oxysporum* f. sp. *ciceri* in seeds shows that the pathogen survived in seeds of all susceptible varieties/ germplasms from 10 to 15 months. Whereas the resistant varieties Phule G-5 and RSG-895 carried the pathogen for a period of 3 and 5 months, respectively. There was prolonged survival of the pathogen was recorded in the variety/ germplasm Radhey, JG-62 and ICC-1375. The pathogen was survived for fifteen months in JG-62, fourteen months in Radhey and twelve months in ICC-1375. The earlier worker have also reported that the pathogen was survived in seeds and other plant parts of the chickpea, but not in pods and leaves (Gangwar *et al.*, 2013; Khune and Patil, 1992).

The results of poly bag experiment presented in Table 3 showed that pre and post-emergence losses due to *F. oxysporum* f. sp. *ciceri* were recorded in the range of 7.0–12.0 per cent and 6.0–10.0 per cent, respectively in different susceptible varieties/ germplasms. The resistant varieties RSG-895 and Phule G-5 showed pre-emergence losses by 3.0 and 2.0 per cent, respectively. Whereas these resistant varieties were not showed the post-emergence losses. The pre and post-emergence losses recorded maximum in the susceptible variety JG-62 followed by Radhey, ICC-1876, ICP-1454, ICC-1375 and DGM-1460. The typical wilt symptoms were developed after 18–22 days of sowing in different susceptible

varieties/ germplasms. They developed earlier on 18th day in the seedlings of JG-62, while they observed delayed on 22nd day in the varieties ICC-1375 and DGM-1460. The seed infection and seed transmission ratio ranged from 3.73:1 to 10.67:1. The maximum seed infection and seed transmission ratio (3.73:1) was found in the variety JG-62, whereas it was minimum (10.67:1) in the variety DGM-1460. It means one seedling showing disease symptom, if we sown 3.73 infected seeds of JG-62 and 10.67 infected seeds of DGM-1460. The seedlings of resistant varieties RSG-895 and Phule G-5 were not showed the disease symptoms.

Table.1 Recovery and per cent seed infection of *F. oxysporum* f. sp. *ciceri* in different varieties/ germplasms in seed washing test, standard blotter method and agar plate method

S. No.	Seed Samples	Recovery of the pathogen in Seed Washing Test	Per cent seed infection in Standard Blotter Method		Per cent seed infection in Agar Plate Method	
			Untreated	**Treated with NaOCl**	**Untreated**	**Treated with NaOCl**
A.	**Resistant varieties/ germplasms**					
1.	***RSG-895***	+*	8.33	4.67	7.00	4.00
2.	***RSG-963***	-	0.00	0.00	0.00	0.00
3.	***RSG-991***	-	0.00	0.00	0.00	0.00
4.	***Phule G-5***	+	6.00	3.33	5.67	3.00
5.	***JG-315***	-	0.00	0.00	0.00	0.00
6.	***BG-373***	-	0.00	0.00	0.00	0.00
B.	**Susceptible varieties/ germplasms**					
7.	***ICC-1375***	+	12.67	7.33	11.33	6.00
8.	***ICC-1876***	+	14.67	9.33	16.33	9.33
9.	***ICP 1454***	+	13.33	10.00	12.00	7.67
10.	***Radhey***	+	16.00	11.33	14.33	9.00
11.	***JG-62***	+	18.67	10.00	17.00	10.67
12.	***DGM-1460***	+	10.67	5.33	8.67	5.00
	Mean		8.36	5.11	7.69	4.55
	SD		6.99	4.44	6.60	4.02
	± SEm		2.02	1.28	1.91	1.16

*+ = Presence of the micro/macro-conidia and mycelial fragments and

– = Absence of the micro/macro-conidia and mycelial fragments of *F. oxysporum* f. sp. *ciceri.*

Table.2 Per cent infection of *F. oxysporum* f. sp. *ciceri* recorded in component plating method and seedling symptom test and duration of its survival in the seeds of different varieties/ germplasms

S. No.	Seed Samples	Per cent infection in different Seed Components			Per cent infection in Seedling Symptom Test			Survival of the pathogen in Seeds (months)
		Seed coat	Cotyledon	Embryonal axis	Initial Per cent of Seed infection	Per cent Seedling Showing Symptom	Seed Infection: Seed Transmission	
A.	**Resistant varieties/ germplasms**							
1.	RSG-895	4.00	0.00	0.00	4.00	0.00	-	5
2.	Phule G-5	0.00	0.00	0.00	4.00	0.00	-	3
B.	**Susceptible varieties/ germplasms**							
3.	***ICC-1375***	14.00	6.00	2.00	8.00	2.00	4.00:1	12
4.	***ICC-1876***	14.00	4.00	2.00	10.00	2.00	5.00:1	10
5.	***ICP 1454***	16.00	6.00	2.00	8.00	4.00	2.00:1	11
6.	***Radhey***	18.00	8.00	4.00	12.00	4.00	3.00:1	14
7.	***JG-62***	20.00	8.00	6.00	14.00	6.00	2.33:1	15
8.	***DGM-1460***	10.00	2.00	0.00	6.00	2.00	3.00:1	10
	Mean	12.00	4.25	2.00	8.25	2.50	3.22:1	10.00
	SD	6.93	3.28	2.14	3.62	2.07	-	4.14
	± SEm	2.45	1.16	0.76	1.28	0.73	-	1.46

Table.3 Pre and post-emergence losses due to *F. oxysporum* f. sp. *ciceri* and parameters related to the disease development in poly bags under field conditions

S. No.	Seed samples	Per cent seed infection recorded in untreated seed in SBM*	Per cent pre-emergence losses	Per cent post-emergence losses	No. of seedlings showing symptoms	Symptoms developed in DAS** (in days)	Seed infection: Seed transmission
A.	**Resistant varieties/ germplasms**						
1.	RSG-895	8.33	3.00	0.00	0.00	0.00	-
2.	Phule G-5	6.00	2.00	0.00	0.00	0.00	-
B.	**Susceptible varieties/ germplasms**						
3.	***ICC-1375***	12.67	7.00	6.00	2.00	22.00	6.33:1
4.	***ICC-1876***	14.67	8.00	7.00	2.00	20.00	7.33:1
5.	***ICP 1454***	13.33	8.00	6.00	3.00	21.00	4.44:1
6.	***Radhey***	16.00	9.00	9.00	4.00	19.00	4.00:1
7.	***JG-62***	18.67	12.00	10.00	5.00	18.00	3.73:1
8.	***DGM-1460***	10.67	7.00	6.00	1.00	22.00	10.67:1
	Mean	12.54	7.00	5.50	2.12	15.25	6.08:1
	SD	4.12	3.21	3.70	1.81	9.51	-
	± SEm	1.46	1.13	1.31	0.64	3.36	-

*SBM = Standard Blotter Method, **DAS = Days After Sowing

The earlier workers have also reported the seed- borne nature of the pathogen (Gangwar *et al*., 2013, Nene *et al*., 1996).

It is concluded that the chickpea wilt caused by *Fusarium oxysporum* f. sp. *ciceri* is a permanent threat to the successful cultivation of the crop. This study throws light on the seed-borne nature of the pathogen. The results of the study showed that the pathogen was carried with the seeds of chickpea, externally on seed surface in the form of mycelial fragments, micro and macro-conidia and internally in the tissues of seed coat, cotyledons and embryonal axis. This finding may help the farmer community to manage the disease and crop more wisely. This study may also support further research regarding the factors which interfere with the cultivation of the crop for the betterment.

References

Anonymous. 2016. Agriculture Statistics at a Glance. Directorate of Economics and Statistics, Department of Agriculture, Cooperation and Farmer Welfare, Ministry of Agriculture and Farmer Welfare, Govt. of India, pp.109-111.

Conci, C., A. Vasquez and S.F. None. 1985. Study of the form of transmission of *Fusarium oxysporum* f. sp. *ciceri* on chickpea. *Phytopathologia*, 20: 57-64.

Gangwar, R.K., R.K. Prajapati and S.R. Singh. 2013. Role of seed and plant debris in the

development of chickpea wilt caused by *Fusarium oxysporum* f. sp. *ciceri*. *Progressive Research*, 8(Sp.) 131-138.

Haware, M.P., Y.L. Nene and R. Rajeswari. 1978. Eradication of *Fusarium oxysporum* f. sp. *ciceri* transmitted in chickpea seed. *Phytopathology*, 68: 1364-1367.

Haware, M.P., Y.L. Nene and N. Natarajan. 1996. The survival of *Fusarium oxysporum* f. sp. *ciceri* in the soil in absence of chickpea. *Phytopathologia Mediterranea*, 35: 9-12.

International Seed Testing Association. 1999. International Rules for Seed Testing. *Seed Science and Technology*, 27: 332.

Khare, M.N., S.B. Mathur and P. Neergaard. 1977. A seedling symptom test for detection of *Septoria nodorum* in wheat seed. *Seed Science and Technology*, 5: 613-617.

Khune, N.N. and M.J. Patil. 1992. Transmission studies of Fusarium wilt by isolation of *Fusarium oxysporum* f. sp. *ciceri* from various plant parts of chickpea. *New Agriculturist*, 3: 227-228.

Kunwar, I.K., K. Satyaprasad and P. Ramarao. 1989. Histopathology of chickpea plant infected with *Fusarium oxysporum* f. sp. *ciceri*. *International Chickpea Newsletter*, 20: 17-18.

Maden, S., D. Singh, S.B. Mathur and P. Neergaard. 1975. Detection and location of seed borne inoculum of *Ascochyta rabiei* and its transmission in chickpea (*Cicer arietinum*). *Seed Science and Technology*, 3: 667-681.

Nema, K.G. and M.N. Khare. 1973. A conspectus of wilt of Bengal gram in Madhya Pradesh Proceeding on "Symposium on wilt problem and breeding for wilt resistance in Bengal gram" helded at IARI, New Delhi, India during September 1973 (Abstr.). pp. 4.

Nene, Y.L., J. Kannaiyan, M.P. Haware and M.V. Reddy. 1979. Review the work done at ICRISAT on soil-borne disease of pigeonpea and chickpea. In proceedings consultants group discussion on the resistance to soil-borne diseases of legumes. ICRISAT, pp. 1-47.

Nene, Y.L., V.K. Sheila and S.B. Sharma. 1996. A world list of chickpea and pigeonpea pathogens. (V Edition), ICRISAT, Patancheru (A.P.) India, pp. 1-27.

Neergaard, P. 1979. *Seed Pathology*. The Mac Millon Press Ltd. London. Vol. I, pp. 835.

Pande, S., J. Narayana Rao and M. Sharma. 2007. Establishment of the chickpea wilt pathogen *F. oxysporum* f. sp. *ciceri* in the soil through seed transmission. *Plant Pathology Journal*, 23: 3-6.

Singh, K.B., S. Weigand, M.P. Haware, Di Vito, M. Malhotra, R.S. Tehan, O. Saxena and L. Holly. 1989. Evaluation of wild species to biotic and abiotic stresses in chickpea (Abst.) XII. Eucarpia Congress, 27 February to 4 March 1989.

Singh, Vikram D. and R.K. Gangwar. 2017. Studies on the seed transmission and management of *Fusarium oxysporum* f. sp. *ciceri* causing chickpea wilt. *International Journal of Innovation Sciences and Research*, 6(7): 1068-1078.

Singh, V.K. 2014. Detection of mycoflora associated with *Cicer arietinum* seeds by agar plate method with PDA. *Weekly Science Research Journal*, 1(30): 1-4.

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