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RESEARCH ARTICLE

Survey of root diseases of chickpea in Jalana district of Marathwada region

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ABSTRACT

A field survey was conducted during 2008-2009, which revealed 5.53 to 11.69 per cent wilt disease in Jalana district of Marathwada region. Survey and surveillance of chickpea wilt in the Jalana district revealed average wilt complex to the tune of 8.43 per cent. Tahsil survey report indicated maximum wilt increase in tahsil Partur (11.69%) followed by Ghansawangi (10.31%), Jalana (10.22%) Bhokardan (9.91 %) Badanapur (7.10%), Ambad (6.77%), Mantha (6.15%) and Jafarabad (5.53%). Further study indicated that *Fusarium oxysporum* f.sp. *ciceri* was associated in majority of cases, pathogen was isolated, purified and its pathogenecity was proved in plastic cup pot. On the basis of morphological, cultural characteristics of pathogen and symptomatology, the fungal pathogen was identified as *a Fusarium oxysporium* f.sp. *ciceri*.

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INTRODUCTION

Chickpea (*Cicer arientinum* L.) is one of the important *Rabi* grain legume cultivated over an area of 4.01 lakh per hectare with the production of 3.11 lakhs tones in Marathwada (Annonymous, 2010). The chickpea wilt caused by *Fusarium oxysporium* f.sp. *ciceri* (Padwick) Snyd. and Hans. is wide spreading almost all the chickpea growing region in the state. The fungus is soil and seed borne and survives in soil in the absence of host or at least 6 year (Haware *et al.* 1986 a and b) causing losses up to 100 per cent. There is an increasing trend in occurrence of the disease in the state due to cultivation of chickpea under irrigated conditions. Considering the nature of damage and survival ability of the fungus, use of resistant varieties is the only economical and practical solution. Most of the resistant varieties have been found to be susceptible after some years because of breakdown in their resistance and evolution of variability in pathogen. Considering the variable types of the wilt reactions of released variety in the farmers field and sick plot at different locations and yield losses caused, the present investigation was undertaken to find out the major causal organisms involved in chickpea wilt complex in Marathwada region of Maharashtra state. Survey and surveillance of chickpea wilt complex incidence of farmer's field was made and collection. isolation, purification and pathogenecity of wilt pathogen were done accordingly.

MATERIALS AND METHODS

Survey and surveillance :

A roving survey of chickpea fields was conducted in tahsils *viz.,* Jalana, Mantha, Partur, Ambad, Ghansawangi, Badnapur, Bhokardan and Jafrabad of Jalana district during the month of December to record the occurrence and distribution of chickpea wilt. On an average, ten farmers' fields of chickpea in each tahsil were visited and the per cent wilt incidence was recorded. Chickpea plants showing typical wilt symptoms were collected in separate paper bag and brought to the laboratory for further investigations.

Isolation, pathogenicity, reisolation and symptomatology :

Chickpea plants, naturally infected and wilted with typical symptoms of wilt were collected from farmer's field and brought to the laboratory. All samples collected from different locations were subjected to the isolation on PDA in the laboratory.

Pathogenicity :

Pathogenicity of the organism was confirmed by sick soil inoculation in plastic cups under green house conditions by using susceptible cultivar, JG-62. The culture of *Fusarium oxysporum* f.sp.*ciceri* was multiplied on Sand maize flour medium. Fifteen gram of maize flour was mixed in 85 gram of river bed sand and filled in the conical flasks of 250 ml capacity (50g / flask) and steriled in autoclave at 1.04 kg cm^2 for 30 minutes. Then these flasks were inoculated aseptically with pure culture of *Fusarium oxysporum* f.sp.*ciceri* and incubated at room temperature for 15 days. After 15 days of incubation, the inoculum was taken out from the flask and mixed thoroughly with sterilized sand + soil mixture (1:1) @ 100 g inoculum per kg soil. This potting mixture (sand + soil+ inoculum) was filled in each plastic cup sterilized with 0.1 % $HgCl_2$ and incubated for four days. Then the seeds of highly susceptible variety, JG-62 were sown @ 10 seeds per plastic cup. The plastic cup with uninoculated soil served as control. All these plastic cup were then watered lightly and kept in glass house for further recording of observations on per cent seed germination, seedling mortality etc. The observations on wilt incidence were recorded after 15 days sowing up to wilting. Re-isolation of the fungus was made from roots artificially inoculated and diseased plants showing the typical symptoms of wilting.

The fungus growth obtained was transferred on PDA slants for comparison with original culture of *F. oxysporum* f.sp. *ciceri*. The symptoms of wilting were observed and recorded right from the initiation of disease till complete wilting of plants both in plastic cup culture as well as field condition. The culture of the pathogen obtained was identified on the basis of morphological and cultural characteristics.

Influence of different media :

Growth characters and sporulation ability of the isolated *Fusarium oxysporum* f.sp.*ciceri* were studied by growing it on different agar culture media. The media used were Czpek's dox agar medium, Asthana and Hawker's medium, Martin Rose Bengal agar, Potato dextrose agar, and Richard's agar. These agar media were prepared by following standard laboratory procedure given by Twite (1969), sterilized by autoclaving and poured in the sterilized Petri plates (ten plates of each medium) allowed to cool down and solidify. Then these plates were inoculated by placing a fungal disc (5 mm diameter) at the centre of the medium in plates and incubated at room temperature for a week.

RESULTS AND DISCUSSION

The results of the present study as well as relevant discussions habe been presented under following sub heads:

Survey and surveillance :

Data are presented in Tables 1 and 2. From the results presented in Table 1, it is revealed that heavy disease incidence was noticed in Partur tahsil (11.69 per cent) followed by Ghansawangi (10.31%) and moderate in Jalna (10.22%) and Bhokardan (9.91%) tahsils. Lowest disease incidence was in Mantha (6.15%) and Jafrabad (5.53%) and average disease incidence was recorded in Jalna district which was to the tune of 8.43 per cent. Similar findings were reported previously by Kohire *et al.* (2006).

Data of Table 2 on per cent wilt incidence in different chickpea cultivars grown on farmers field indicated, maximum incidence of wilt (average 12.82%) in local chickpea cultivar followed by BDN-9-3(7.95%) and Vishal (7.65%) and lowest incidence (3.78 %) was recorded in BDNG-797. Tahsil wise wilt incidence revealed that in all cultivars including local, maximum incidence was recorded in tahsil 16.25 per cent followed by Partur (15.25%) and Jalna (15.03%).

Results presented in Table 3 revealed that out of 80 samples isolated and studied, near about 67 (77.90%) proved the association of *Fusarium oxysporum* f.sp. *ciceri* followed by 15 (17.44%) of *Rhizoctonia bataticola* and 4 (4.65%) of *Sclerotium rolfsii* with chickpea wilt complex. Kohire *et al.* (2006) carried out survey and surveillance of chickpea wilt and reported that disease incidence of wilt varied from 6.6 to 18.5 per cent. These results clearly indicated that the major pathogen associated with chickpea wilt complex was *Fusarium oxysoprum* f.s. *ciceri* during early as well as later stages of the crop and to some extent, *Rhizoctonia bataticola* and *Sclerotium rolfsii* specially in early stage of the crop.

Isolation and pathogenicity :

The fungus, *Fusarium oxysporum* f.sp. *ciceri* was isolated from the wilted plants collected during the survey of Jalna district. The pure culture was obtained by hyphal tip method, subcultured frequently and maintained PDA slants for further studies.

Pathogenicity of the fungus was carried out in plastic cup by soil inoculation method using variety JG-62 which exhibited wilting after 30 days of inoculation. The findings of this test are presented in Table 4 and 5 . The inoculated seeds with pathogen, *Fusarium oxysporum* f. sp. *ciceri* exhibited 65 per cent overall germination where 40 per cent of the population mortality observed in pre-emergence condition and 21 per cent and 72 per cent post-emergence mortality had been occurred on 2nd and 3rd week after inoculation, respectively (Table 4). Similarly, the leaf yellowing of 60 per cent and 75 per cent was observed on 3rd and 5th day after inoculation, respectively with 100 per cent seedling mortality (Table 5).

Symptoms of wilting produced by artificially inoculated and diseased plants were identical and confirmed with those

Table 1 : Survey of different villages for incidence of chickpea wilt at flowering / podding stage under irrigated / rainfed conditions (%)

Tahsils of Jalna district	Place	No. of samples collected	Soil type	Type of farming	Stage of crop	Samples per cent incidence					Average incidence (%)	Range incidence (%)
						I	II	III	IV	V		
Jalna	Golapangari, Ramnagar, Ner	12	Medium black	Irrigated	Flowering	10.56 (18.91)	9.25 (17.66)	8.20 (16.64)	10.50 (18.91)	12.62 (20.79)	10.22 (18.63)	8.20 to 12.62
Mantha	Kendhali, Watur, Akani	12	Light	Rainfed	Podding	8.60 (17.05)	6.50 (14.77)	6.35 (14.54)	3.20 (10.31)	6.12 (14.30)	6.15 (14.30)	3.20 to 8.60
Partur	Satona, Partur, Rohina	10	Light	Rainfed	Flowering	14.30 (22.22)	16.80 (24.20)	11.90 (20.18)	9.95 (18.34)	5.54 (13.56)	11.69 (19.91)	5.54 to 16.80
Ambad	Shahagad, Wadi godari, Hadgaon	12	Black	Irrigated	Podding	4.33 (18.97)	8.25 (16.64)	10.80 (19.19)	5.50 (13.56)	5.00 (12.92)	6.77 (15.00)	4.33 to 12.80
Ghansawangi	Rajegaon, Ku. Pimplegaon, Bodkha	12	Black	Irrigated	Flowering	10.25 (18.63)	12.50 (20.70)	9.80 (18.24)	10.25 (18.63)	8.75 (17.16)	10.31 (18.72)	8.75 to 12.50
Badnapur	Selgaon, Kandari, Ambadgaon	10	Medium black	Renfed	Podding	6.06 (14.18)	9.80 (18.24)	5.90 (14.06)	6.25 (14.42)	7.50 (15.89)	7.10 (15.45)	5.90 to 9.80
Bhokardhan	Paradh, Bhokardan, Virogaon	9	Light	Rainfed	Flowering	9.10 (17.56)	11.90 (20.18)	12.25 (20.44)	7.50 (15.89)	8.80 (17.26)	9.91 (18.34)	7.50 to 11.90
Jafrabad	Tembhurni, Mahhura, Deogaon	7	Light	Rainfed	Podding	6.25 (14.42)	7.50 (15.89)	5.25 (13.18)	4.75 (12.52)	3.90 (11.39)	5.53 (13.56)	3.90 to 7.50
S.E.											1.67	
C.D. at 5%											5.01	
C.V. %											23.35	

The figure sin parentheses are arsing values

symptoms observed on naturally infected and wilted chickpea plants in the field. The symptoms produced were exactly identical to those described earlier by Shinde (2003) and Haware *et al.* (1986b). The morphological and cultural characteristics of *Fusarium oxysporum* f.sp. *ciceri* obtained after re-isolation were similar to those reported earlier by several workers Gupta *et al.,* 1986; Nikam *et al.,* 2008. Pure culture of *Fusarium oxysporum* f.sp. *ciceri* was again reinoculated on chickpea cultivar JG-62 in plastic cup and the symptoms appeared were drooping of petioles and rachis along with leaflets. Such seedlings further got collapsed retaining almost their green colour but when uprooted showed uneven shrinking of stem above and below the collar region. These symptoms of chickpea wilt observed were similar to

Table 2 : Per cent wilt incidence in chickpea cultivars on farmers field

Name of tahsil	No. of locations traversed	Per cent wilt				
		Vijay	BDN 9-3	Vishal	BDNG 797	Local
Jalna	15	5.8	6.0	7.6	3.5	15.30
Mantha	10	9.2	7.5	7.7	5.0	16.25
Partur	9	7.8	8.8	9.2	4.1	15.25
Ambad	10	6.6	7.5	8.5	2.5	14.75
Ghansawangi	12	8.1	9.7	7.8	1.8	12.80
Badnapur	9	7.5	8.5	6.7	5.2	12.40
Bhokardhan	7	7.0	6.8	7.2	2.7	11.60
Jafrabad	7	7.2	8.8	6.5	4.5	10.5
Average		7.4	7.95	7.65	3.78	12.82

Table 3 : Study of organisms associated with chickpea wilt complex

Name of tahsil	Number of samples collected	Type of pathogen isolated		
		Fusarium oxysporum f. sp.ciceri	*Rhizoctonia bataticola*	*Sclerotium rolfsii*
Jalna	10	9	1	1
Mantha	10	8	2	0
Partur	10	8	3	0
Ambad	10	10	1	1
Ghansawangi	10	8	3	1
Badnapur	10	8	3	1
Bhokardhan	10	9	0	0
Jafrabad	10	7	2	0
Total	80	67 (77.90)	15 (17.44)	4 (4.65)
Average	10	8.3	1.8	0.5

Table 4 : Pathogencity test of wilt pathogen (*Fusarium oxysporum* f. sp. *ciceri*) by soil inoculation method

Particulars	*Germination (%)	*Pre-emergence mortality (%)	*Post-emergence mortality (%)	
			2nd week	3rd week
Inoculated	65.00	40.00	21.00	72.00
Control	100.00	0.00	0.00	0.00

*Average of three replications

Table 5 : Pathogencity test of wilt pathogen (*Fusarium oxysporum* f. sp. *ciceri*) by soil inoculation method

Particulars	*Leaf yellowing (%)		Seedling mortality (%)
	3 DAI	5 DAI	
Inoculated	60.00	75.00	100.00
Control	0.00	0.00	0.00

DAI – Days after inoculation

*Average of three replications

Table 6 : Influence of different culture media on radial growth (mm) and sporulation of wilt pathogen (*Fusarium oxysporum* f. sp. *ciceri*)

Media	**Growth (mm)			*Sporulation
	5 DAI	7 DAI	9 DAI	
Czapek's dox agar medium	35.00	44.80	64.90	26.80
Asthana and Hawker's medium	40.30	60.25	72.75	28.25
Martin's Rose Bengal agar	20.66	25.35	30.25	20.30
Potato dextrose agar	52.25	59.05	79.25	39.60
Richard's agar	25.35	31.30	39.05	19.40
Mean	37.71	44.15	57.24	26.87
S.E. $\pm$	0.891	1.65	1.28	
C.D. at 5%	2.67	4.95	3.84	

**Average sporulation per microscopic field *Average of three replication DAI – Days after inoculation

those recorded by Khillare *et al.* (2007) and Kewate (1986).

Identification of fungal pathogen :

The pathogenic cultures isolated from diseased plants were identified on the basis of morphological characters as *Fusarium oxysporum* f.sp. *ciceri* and confirmed from Department of Mycology and Plant Pathology, I.A.R.I., New Delhi.

Influence of different culture media :

The results in respect of growth characters of *Fusarium oxysporum* f.sp. *ciceri* on various culture media are presented in Table 6. From the result, it is revealed that maximum growth was obtained on Potato dextrose agar medium which was found significantly superior for radial growth and sporulation of wilt pathogen over rest of the medium tested. The Martin Rose Bengal agar medium recorded significantly lower radial growth at fifth (20.66 mm), seventh (25.35 mm) and ninth (30.25 mm) days after inoculation as compared to rest of media. The sporulation of *F. oxysporum* f.sp. *ciceri* was good on PDA medium (39.60 mm) followed by Asthana and Hawker's medium (28.25mm) and Czapek's dox agar medium (26.80 mm). These results obtained are in agreement with the findings of Khillare *et al.* (2007) and Osman *et al.* (1992) who reported Potato extract agar as a best medium for mycelial growth and sporulation of *F. oxysporum* f.sp. *ciceri.*

REFERENCES

Gupta, O., Khare, M.N. and Kotushane, S.R. (1986). Variability among six isolates of *Fusarium oxysporum* f.sp. *ciceri* causing vascular wilt of chickpea. *Indian Phytopathol.*, **39**: 279-281.

Haware, M.P., Nene, Y.L. and Mathure, S.B. (1986a). Seed borne diseases of chickpea. Technical Bulletin 1. Danish Government Institute of Seed Pathology for Developing Countries. *Copenhagen,* **1**: 1-32.

Haware, M.P., Nene, Y.L. and Natrajan, M. (1986b). Survival of *Fusarium oxysporum* f.sp. *ciceri* in soil in absence of chickpea. Paper presented in the National seminar on management of soil borne diseases of crop plants. Proc. Natn. Sem. 8-10, Jan. 1986. Tamil Nadu Agriculture University, Coimbatore (T.N.) INDIA.

Kewate, S.B. (1986). Studies on wilt of gram (*Cicer arietinum* L.) caused by *Fusarium oxysporum* f.sp. *ciceri*. M.Sc.(Ag.) Thesis, Mahatama Phule Krishi Vidyapeeth Rahuri, Ahmednagar, M.S. (INDIA).

Khillare, V.C., Rafi Ahmed and Kohire, O.D. (2007). Characterization *of Fusarium oxysporum* f.sp. *ciceri* causing chickpea wilt in Maharashtra. Paper presented in 2nd Asian Congress of Mycology and Plant Pathology held at Hyderabad on 19-22, Dec. 2007, p.65.

Kohire, O.D., Shrirame, M.D., Dahiwal, A.L., Shrirarkar, P.D. and Vishnupulikar, R.M. (2006). Status of chickpea wilt dry root rot and stunt in major chickpea growing district of Marathwada region of Maharashtra. Paper presented in Agro-research Conservation and Management held at Raipur on 14-15 Dec. 2006.

Nikam, P.S., Jagtap, G.P. and Bidwe, K.U. (2008). Survey and surveillance of chickpea wilt complex caused by *Fusarium oxysporum* f. sp. *ciceri. J. pl. Disease Sci.,* **3**(1):29-33.

Osman, M., Sayed, M.A., Mohamed, Y.A.H. and Metwally, M. (1992). Effect of various cultural conditions on *Alternaria alternata* and *Fusarium oxysporum* I. Culture media, temperature, age and carbon source. *Microbios.*, **71** (286): 15-26.

Shinde, V.S. (2003). Studies on wilt of chickpea (*Cicer aretinum* L.). M.Sc.(Ag.) Thesis, Mahatama Phule Krishi Vidyapeeth Rahuri, M.S. (INDIA).

Twite, J. (1969). *Plant Pathological methods* : *Fungi and bacteria*. Burgess Publishing Co., pp. 12-80.

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