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Short Note

Pathogenicity and Morphological Variabilities of *Fusarium oxysporum* f.sp. *cepae* Isolates in Onion

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Basal rot disease infected onion bulbs were collected from fifteen places of Tamil Nadu, India. Among the fifteen isolates screened for virulence, DNSFOC1 isolate collected from Sempatti in Dindugal district recorded maximum (88.45) per cent disease infection under artificial inoculation. DNSFOC1 significantly recorded 29.93 mm mycelial growth per day. The length of macroconidia varied from 20.25 to 27.43 µm and the width ranged from 2.36 to 3.16 µm. The length of microconidia varied among the isolates from 7.10 to 8.43 µm and the width ranged from 2.14 to 2.67 µm. Compact, fluffy and sparse colony types were observed among the isolates. Significant variation was observed in morphological character and virulence among the *Fusarium oxysporum* f. sp. *cepae* isolates.

Key words: onion, morphological character, pathogenicity, *Fusarium oxysporum* f. sp. *cepae*

Onion (*Allium cepa* var *aggregatum* G.Don) is one of the important vegetable crops grown in India. China ranks first in world onion production followed by India, USA, Turkey, Pakistan, Russia, Indonesia, Vietnam, and Myanmar. Basal rot is a devastating disease of onion caused by *Fusarium oxysporum* Schlechtend: Fr. f. sp. *cepae* (Hans.) (Coskuntuna and Ozer, 2008). In India the incidence of basal rot was first reported by Mathur and Shukla (1963). In Tamil Nadu, this disease was first observed by Ramakrishnan and Eswaramoorthy (1982) from Coimbatore district. The fungus attacks seedlings, causing pre and post emergence damping off, root rot of older plants, and stem plate discoloration and basal rot of bulbs in the field and in storage (Abawi and Lorbeer, 1972). It can significantly reduce the crop yield to approximately 45 t/ha in USA (Schwartz *et al*., 1991). In Japan, during summer more than 50 per cent loss occurs in welsh onion due to *Fusarium* basal rot (Dissanayake *et al*., 2009).

Morphological characters are important tools in the identification and classification of fungus. Several workers described enormous pathogenic variability among *Fusarium oxysporum* f.sp. *pisi* isolates (Whitehead *et al*., 1992; Kraft, 1994). Singh *et al*., (2010) studied the variability in cultural characteristics and virulence among three isolates of *Fusarium oxysporum* f.sp. *ciceri* in chickpea under *in vitro* condition. Hence, the present investigation was conducted to study the morphological and pathogenic variability among isolates of *Fusarium oxysporum* f. sp. *cepae*.

Materials and Methods

Isolation of pathogen

Basal rot disease infected onion bulbs were collected from onion growing areas of Tamil Nadu, India during 2008-2009. The pathogen was isolated from the diseased tissues of onion by tissue segment method (Rangaswami, 1958). Infected portions of diseased plants were cut into small pieces using sterilized scalpel and surface sterilized with 0.1 per cent mercuric chloride for one minute and washed in three changes of sterile distilled water and then placed on Petri dish containing solidified Potato Dextrose Agar (PDA) medium. These plates were incubated at room temperature (28 ± 2ºC) for five days and observed for the growth of the fungus. The hyphal tips of fungi were transferred aseptically to PDA slants for maintenance of the culture.

Morphological variability among Fusarium oxysporum f. sp. cepae isolates

From the five day old culture plates nine mm culture disc of the pathogen was cut by a sterilized cork borer and placed at the center of each sterile Petri dish containing 20 ml of previously sterilized and solidified PDA medium. The plates were incubated at room temperature (28+2$_0$C) for five days. The growth and morphological characters of the isolates *viz*., colony morphology, mycelial growth rate, colony colour, conidia size, shape and septation were observed, measurement was made under the microscope (Olympus BX41).

Pathogenic variability among Fusarium oxysporum f. sp. cepae isolates

The isolates of *Fusarium oxysporum* f. sp. *cepae* were multiplied on sand-maize medium (Riker and Riker, 1936). The medium containing sand and

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maize powder (19:1) was mixed, moistened with 400 ml of water kg^{-1} and then packed in polypropylene bags. The bags were sterilized in an autoclave at 120^0C with 15 psi for 20 minutes and continued the sterilization for two concecutive days. Two nine mm PDA culture disc of actively growing *Fusarium oxysporum* f. sp*. cepae* were inoculated in each polypropylene bag replicated three times. They were incubated at room temperature (28±2°C) for 15 days and used as source of inoculum

Virulence of isolates of *Fusarium oxysporum* f. sp*. cepae*

Earthen pots of uniform size (30 cm diameter) were filled with five kg of sterilized garden land soil. Five gram inoculum of each isolates (multiplied on sand maize medium) of the pathogen was mixed with soil present in pots. Four onion bulbs (cv. Co-5) were sown in each pot and replicated three times. The pots were maintained in green house by regular, uniform and careful watering and the growth was constantly observed for development of the disease symptoms. The per cent disease incidence of each isolate was recorded after 25 days of inoculation.

Results and Discussion

The mycelial growth habit of fifteen isolates was studied of which isolate DNSFOC1 significantly recorded 29.93 mm mycelial growth per day followed by ERVFOC2 which recorded 26.45 mm growth per day(Table 1). Minimum mycelia growth was observed in COAFOC3 (21.45 mm) isolate. The mycelial colours of isolates were light brown, dark brown, light yellow and pink. Compact, fluffy and sparse colony types were observed among the isolates. *Fusarium oxysporum* f. sp*. ciceri* isolates (Foc1, Foc2, Foc3) showed dull white, flat hairy growth and white fluffy colony with irregular margin (Singh *et al.,* 2010)

Table 1. Morphological characters of isolates of *Fusarium oxysporum* f. sp. *cepae*

Isolates	Place of collection	Colony type	Colour of culture medium	Growth rate (mm/day)*	Macro conidia		Micro conidia	
					Septation	Size (µm)	Septation	Size (µm)
ERKFOC1	Kodumudi	Compact	Light brown	23.23	2-3	24.45×3.12	0	7.10×2.52
ERVFOC2	Vengumpur	Sparse	Dark brown	26.45	2-3	22.45×3.05	0	7.82×2.32
ERPFOC3	Perumalkovil pudhur	Fluffy	Light brown	25.27	2-3	20.34×2.43	0	7.32×2.24
MDSFOC1	Sekkanurani	Compact	Light yellow	23.85	3-4	22.46×2.87	0	7.61×2.27
MDPFOC2	Palamedu	Fluffy	Dark brown	25.42	3-4	23.38×2.45	0	7.52×2.31
MDUFOC3	Usilampatti	Sparse	Light brown	24.54	2-3	22.35×2.57	0	7.94×2.52
DNSFOC1	Sempatti	Fluffy	Pink	29.93	2-3	27.43×3.16	0	8.43×2.67
DNOFOC2	Ottanchathram	Compact	Light yellow	26.21	2-3	23.57×2.45	0	7.82×2.32
DNTFOC3	Tharmathupatti	Fluffy	Dark brown	22.91	2-3	25.49×2.63	0	7.32×2.14
COUFOC1	Udumalpet	Compact	Light brown	21.56	3-4	22.16×2.45	0	7.61×2.27
COPFOC2	Pedhappampatti	Compact	Light yellow	24.82	3-4	23.65×2.18	0	7.52×2.31
COAFOC3	Annur	Sparse	Dark brown	21.45	2-3	24.38×2.72	0	7.83×2.52
THMFOC1	Mettupatti	Fluffy	Light brown	23.94	2-3	24.31×2.61	0	8.21×2.54
THBFOC2	Bodinayakanur	Compact	Light yellow	24.57	2-3	23.45×2.36	0	7.42×2.21
THPFOC3	Periyakulam	Sparse	Dark brown	21.59	2-3	20.25×2.91	0	7.73×2.42
CD(P=0.05)				2.04				

*Mean of three replications

Fusarium oxysporum f. sp*. cepae* produced macroconidia and microconidia. The microconidia were oval to reniform while the macroconidia were falcate to straight, apical cell somewhat pointed. The septation of the macroconidia varied from 2-4 among the isolates. The length of macroconidia varied from 20.25 to 27.43 µm and the width ranged from 2.36 to 3.16 µm (Table 1). The maximum conidial length and width were observed in DNSFOC1 (27.43 and 3.16 µm). The minimum conidial length was observed in THPFOC3 (20.25 µm). The minimum conidial width was observed in THBFOC2 (2.36 µm). *Fusarium oxysporum* f. sp*. ciceri* (foc2) showed 3 to 6 septa in conidia whereas Foc 1 and Foc 3 had 2 to 3 septate (Singh *et al*., 2010).

The length of microconidia varied among the isolates from 7.10 to 8.43 µm and the width ranged from 2.14 to 2.67 µm. Maximum conidial length and width was observed in DNSFOC1 (8.43 and 2.67 µm) and minimum conidial length was observed in ERKFOC1 (7.10 µm). Minimum conidial width was observed in DNTFOC3 (2.14 µm). Alice (1994) reported variation in morphological characters of *Fusarium oxysporum* in onion. Thirty isolates of *Rhizoctonia bataticola* collected from major chickpea growing areas were highly variable in their cultural and morphological characters (Manjunatha and Naik, 2011). The results indicated that difference in morphological character was positively correlated with its virulence.

Per cent disease incidence varied from 32.65 to 88.45. DNSFOC1 isolate collected from Sempatti in Dindugal district recorded 88.45 per cent disease infection under artificial inoculation and was identified as a virulent culture where as minimum

infection recorded in ERPFOC3 isolate (32.65 PDI) (Table 2). DNSFOC1 exhibited yellowing and wilting of leaves within twenty days after inoculation. The leaf tip dieback eventually progressed to the entire leaf and infected onion bulbs were pulled out easily.

Table 2.Virulence of isolates of *Fusarium oxysporum* f. sp. *cepae* on onion in pot culture

Isolates	Place of collection	District	* Basal rot incidence (%)
ERKFOC1	Kodumudi	Erode	76.82 (61.22)**
ERVFOC2	Vengumpur		82.64 (65.37)
ERPFOC3	Perumalkovil pudhur		32.65 (34.84)
MDSFOC1	Sekkanurani	Madurai	62.75 (52.38)
MDPFOC2	Palamedu		54.23 (47.42)
MDUFOC3	Usilampatti		45.78 (42.58)
DNSFOC1	Sempatti	Dindugal	88.45 (70.13)
DNOFOC2	Ottanchathram		75.82 (60.54)
DNTFOC3	Tharmathupatti		65.21 (53.85)
COUFOC1	Udumalpet	Coimbatore	54.20 (47.41)
COPFOC2	Pedhappampatti		62.85 (52.44)
COAFOC3	Annur		47.82 (43.75)
THMFOC1	Mettupatti	Theni	76.21 (60.80)
THBFOC2	Bodinayakanur		79.41 (63.01)
THPFOC3	Periyakulam		66.50 (54.63)
CD(P=0.05)			1.35

*Mean of three locations ** Figures in the parentheses are arc sine transformed values

Jimenez Diaz *et al*. (1993) revealed that isolates of *Fusarium oxysporum* f.sp.*pisi* showed high pathogenic variability which may limit the efficiency of resistance. Diversity in cultural, morphological characters and pathogenicity among six isolates of *Fusarium oxysporum* f.sp.*ciceri* in chickpea was studied by Barhate *et al*. (2006). Morphological and pathogenic variations were observed in the isolates of *Fusarium oxysporum* f.sp. *pisi* collected from different pea growing areas of Himachal Pradesh (Gupta *et al.,* 2011).

The present investigation revealed that the isolates of *Fusarium oxysporum* f. sp. *cepae* exhibited high variability in morphological character and pathogenicity which could be used for further development of race specific resistant varieties of onion for the control of disease.

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