ISSN: 0976-3031

Available Online at http://www.recentscientific.com

CODEN: IJRSFP (USA)

International Journal of Recent Scientific Research
Vol. 8, Issue, 12, pp. 22702-22704, December, 2017

International Journal of Recent Scientific Research

DOI: 10.24327/IJRSR

Research Article

SURVEY AND SEVERITY OF TOMATO WILT DISEASE INCITED BY *FUSARIUM OXYSPORUM* F. SP. *LYCOPERSICI* (SACC.) IN DIFFERENT DISTRICTS OF TAMILNADU

Amutha C and Darwin Christdhas Henry L

Department of Plant Pathology, Faculty of Agriculture, Annamalai University

DOI: http://dx.doi.org/10.24327/ijrsr.2017.0812.1320

ARTICLE INFO

Article History:

Received 05th September, 2017
Received in revised form 21st October, 2017
Accepted 06th November, 2017
Published online 28th December, 2017

Key Words:

Survey, Soil borne pathogen, *Fusarium* wilt, Disease severity, Tomato

ABSTRACT

A survey was conducted to investigate the incidence and severity of tomato wilt caused by *Fusarium oxysporum* f. sp. *lycopersici* in ten major tomato growing districts of TamilNadu. Sivagiri in Tirunelveli district followed by Ponchamalli in Krishnagiri district recorded maximum PDI, whereas Vadakadu in district recorded minimum PDI. Isolates Fol 1 showed maximum growth rate/day followed by isolate Fol 17. Isolate Fol 1 was found to be highly virulent causing 100% wilt at 30 DAS in soil inoculation method followed by isolates Fol 17&Fol 18 in the decreasing order of merit. It is also proved that the variations in morphological characters correlates with the virility of the different isolates.

INTRODUCTION

Tomato (*Lycopersiconesculentum*Mill) is one of the most widely cultivated food crops (Singh & Singh, 2004). In Tamil Nadu, tomato is grown in an area of 22,433 ha, with a production of 2, 82,912 tonnes and a productivity of 12,611 kg/ha. Tomato is attacked by many fungal pathogens in the field and *Fusarium* wilt of tomato caused by the fungal pathogen *Fusarium oxysporum* f. sp. *lycopersici* (Sacc.) is one of the most important diseases, which affect during all the stages of the plant (seedling stage, flowering stage, and fruiting stage). Also, it can affect the whole plant parts, leaves and stems. Fusarium oxysporum f.sp.lycopersiciis a phytopathogenic fungus causing vascular wilt disease on tomato, known as fusariosis (Di Pietro et al., 2003). The disease was more severe when tomato crop was grown as monoculture (Handerson and Winslead, 1961). It causes great losses, especially under favorable weather conditions. Occasionally entire fields of tomatoes are destroyed or severely damaged before a crop can be harvested. The first symptom of *Fusarium* wilt in gardens and fields is usually the golden yellowing of a single leaflet or shoot, or a slight wilting and drooping of the lower leaves on a single stem. Madhavi *et al*., (2006). Leaves or whole branches will turn yellow, then brown and die still attached to the plant described as a yellow-flagging appearance. (Alexander and Tucker, 1945). At advanced stage, browning of the vascular system can be seen and pathogen induces severe wilting of plants by blocking xylem vascular bundles and impending the movement of water (Decal et al., 2000). The fungus invades epidermal tissues of the root, extends to the vascular bundles, produces mycelia and/or spores in the vessels, and results in death of the plants (El – Khallal, 2007). Sometimes half of a leaf or branch will be affected, with the other half seemingly unaffected. The fungus can be observed as brown discoloration in the vascular tissue of affected branches (Agrios, 2005). A survey was undertaken to assess the morphological variations of different isolates of *Fusarium oxysporum* f. sp. *lycopersici,* Percent Disease Incidence in major tomato growing districts of Tirunelveli, Madurai, Salem, Dharmapuri, Ariyalur, Cuddalore, Coimbatore, Pudukkottai, Krishnagiri, Viruthunagar districts in Tamil Nadu and the virulence of certain screened isolates..

MATERIALS AND METHODS

Survey for incidence of tomato wiltat major tomato growing districts of Tamil Nadu Extensive surveys were made on the incidence of tomato wilt disease in 20 villages among 10 districts of Tamil Nadu during post rainy season of 2016. The total number of plants wilted were recorded in one sq. meter of area and the Percent Disease Incidence (PDI) was calculated for each field location by using the formula mentioned below.

Corresponding author:* **Amutha C
Department of Plant Pathology, Faculty of Agriculture, Annamalai University

$$\text{PDI} = \frac{\text{Total number infected plants}}{\text{Total number of plants}} \times 100$$

Isolation and identification of the Pathogen

Affected roots and collar portions of tomato plant showing typical symptoms of wilt disease was collected and the pathogen was isolated and maintained in potato dextrose agar (PDA) medium *in vitro* at 25 ± 2°C for seven days. Pure culture of the pathogen was obtained by single hyphal tip method (Rangaswami, 2005). Identification of *Fusarium* spp. was made on the basis of morphological and cultural characters described by Booth (1971). Following pathogenic isolates of *F. oxysporum* f. sp. *lycopersici* (Fol 1 to Fol 20) were used for various experiments.

Cultural and morphological characters on PDA

Growth diameter of each isolate was measured. The pigmentation was recorded 10 days after inoculation. Microscopic examination was carried out for chlamydospore formation after 25 days of inoculation. Fifty micro conidia as well as macro conidia were measured with stage and ocular micrometers and average values worked out. Sporulation was studied by means of haemocytometer under the microscope. Five discs of fungal growth of 5 mm diameter were dissolved in 20 ml of distilled water and shaken well to get a spore suspension.

Virulence test

Soil inoculation*:* the soil mixture of silt and sand in 3:1 ratio (v:v), respectively, was sterilized at 100ºC for 60 minutes in an autoclave. ***Inoculum*****:** Conidial suspensions were adjusted to concentrations of (100 spore/ml) using haemocytometer. 50 ml spore suspension of the fungus was poured per pot and mixed with the soil. Pots with just sterilized soil served as control. The pots were designed as randomized block in greenhouse bench with air temperature 25-32ºC.

Table 1 Survey on Fusarium wilt disease incidence of tomato in major tomato growing areas of Tamilnadu.

S.No	Districts	Location	Isolates	Variety	Percent disease incidence (PDI)*
1.	Tirunelveli	Sivagiri	Fol 1	PKM1	58.83 (58.45)
2.		Athuvazhi	Fol 2	CO3 (Marutham)	49.45 (48.40)
3.	Madurai	Solavanthan	Fol 3	CO1	40.40 (46.30)
4.		Checkkanrani	Fol 4	COTH3	33.20 (38.45)
5.	Ariyalur	Veerakkan	Fol 5	CO2	43.82 (32.25)
6.		Pillakurichi	Fol 6	COLCRH3	35.75 (43.82)
7.	Pudukkottai	Vadakadu	Fol 7	PKM1	12.65 (34.61)
8.		Kattiyavayal	Fol 8	CO3 (Marutham)	21.78 (35.65)
9.	Cuddalore	Vallampadukai	Fol 9	CO1	32.82 (12.65)
10.		Sivapuri	Fol 10	PAIYUR1	36.75 (21.78)
11.	Virudhunagar	Sethur	Fol 11	CO3 (Marutham	40.21 (18.78)
12.		Nedungkulam	Fol 12	local	17.75 (32.50)
13.	Coimbatore	Perur	Fol 13	CO3 (Marutham)	39.23 (35.70)
14.		Thondamuthur	Fol 14	CO1	41.42 (40.21)
15.	Salem	Athur	Fol 15	PKM1	46.82 (40.18)
16.		Adapady	Fol 16	CO2	36.85 (15.75)
17.	Krishnagiri	Ponchamalli	Fol 17	COLCRH3	55.54 (38.25)
18.		Krishnapuram	Fol 18	Arkavishal	47.21 (41.42)
19.	Dharmapuri	Machery	Fol 19	Arkaabha	34.32 (39.46)
20.		Penagram	Fol 20	Hisarlalima	31.44 (28.41)
CD(P)=0.05					**7.51**

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

Morphological characters of F.o.f.sp .lycopersici isolates.

From the five days old culture plates nine mm culture disc of the pathogen was cut by using a sterilized cork borer and placed at the center of the each sterile Petri dish containing 15 ml of previously sterilized and solidified PDA medium. The plates were incubated at room temperature at (28+2^{o}C) for five days. The growth and morphological characters of the isolates *viz*., colony morphology, mycelial growth rate, colony colour, conidia size, shape and septations were observed, measurements were taken under microscope (magnification 45x) after calibration with ocular and stage micrometer.

RESULTS AND DISCUSSION

The results of the survey revealed that the incidence of wilt disease showed its widespread occurrence in almost all tomato growing areas of Tamilnadu ranging from (12% to 59%). The maximum PDI of 58.83 per cent was recorded at Sivagiri in Tirunelveli district followed by Ponchamalli in Krishnagiri district which recorded 55.54Per cent, where as Vadakadu in Pudukkottai district recorded the minimum of 12.65 PDI. On the average Tirunelveli district followed by Krishnagiri district and Salem district recorded the maximum Per cent disease incidence. Manikandan and Raguchander (2014) also reported that, the incidence of wilt disease was prevalent in almost all tomato growing areas of Tamil Nadu.

Among the 20 isolates tested Fol1 recorded a maximum growth rate of 26.96mm/day followed by Fol17 fromPonchamalli recording 26.90mm/day. The least growth rate was measured by isolate Fol 7 recording 18.45mm/day. Morphological characters are important tools in identification and classification of the fungus. In the present study, spore size, septation of conidia and shape of conidia were used for identifying the fungus. Twenty isolates of F.o. f.sp. lycopersici varied in all the characters. Out of these twenty isolates, eight isolates were fluffy colonies, seven isolates were compact and five isolates produced sparse mycelial growth. Micro conidia were small, oval shaped, hyaline, single or bicelled. Macroconidia were sickle shaped hyaline and multicelled with three to five septations. Similar findings has been reported by Rekah et al., (2000) and Manikandan and Raguchander (2014). The size of the microconidia and macroconidia were found to be maximum in Fol 1 followed by Fol 17 and Fol 2 recording (27.43×3.16 & 8.43×2.67 µm; 26.65×2.18 & 8.43×2.67 µm and 24.31×2.61 & 8.21×2.54 µm) respectively. It was observed that under pot culture experiments, in soil inoculated with the pathogen(Table 3) all the isolates of F. oxysporum f. sp. Lycopersici showed variation in their virulence in causing wilt and among the 10 isolates screened, the isolate Fol 1 was found to be highly virulent causing 100% wilt at 30 DAS in soil inoculation method followed by Fol 17, recording 96 Per

Table 2 Morphological characters of different isolates of F. oxysporum f. sp. lycopersici

S.No	Isolates	Place of collection	Colony type	Colour of the culture medium	Growth rate (mm/day)*	Macro conidia		Micro conidia	
						Septation	Size (µm)	Septation	Size (µm)
1	Fol 1	Sivagiri	Compact	Light brown	26.96	3-4	27.43×3.16	0	8.43×2.67
2	Fol 2	Athuvazhi	Compact	Dark brown	23.45	2-3	24.31×2.61	0	8.21×2.54
3	Fol 3	Solavanthan	Fluffy	Light brown	21.82	2-3	22.34×2.43	0	7.32×2.24
4	Fol 4	Checkkanrani	Compact	Light yellow	20.23	3-4	20.46×2.87	0	7.61×2.27
5	Fol 5	Veerakkan	Fluffy	Dark brown	22.27	2-3	24.43×3.16	0	7.94×2.52
6	Fol 6	Pillakurichi	Fluffy	Light brown	20.54	2-3	22.49×2.63	0	7.82×2.32
7	Fol 7	Vadakadu	Sparse	Pink	18.45	2-3	22.45×3.12	0	7.10×2.52
8	Fol 8	Kattiyavayal	Sparse	Light yellow	18.56	2-3	21.57×2.45	0	7.82×2.32
9	Fol 9	Vallampadukai	Fluffy	Dark brown	18.59	2-3	21.49×2.63	0	7.32×2.14
10	Fol 10	Sivapuri	Compact	Light brown	19.91	3-4	22.16×2.45	0	7.61×2.27
11	Fol 11	Sethur	Compact	Light yellow	21.57	2-3	23.65×2.18	0	7.61×2.27
12	Fol 12	Nedungkulam	Sparse	Dark brown	18.99	2-3	21.45×3.05	0	7.23×2.22
13	Fol 13	Perur	Fluffy	Light brown	21.85	2-3	22.25×2.91	0	7.73×2.42
14	Fol 14	Thondamuthur	Compact	Light yellow	21.01	2-3	20.45×2.36	0	7.42×2.21
15	Fol 15	Athur	Compact	Dark brown	23.21	2-3	23.38×2.72	0	7.82×2.32
16	Fol 16	Adapady	Fluffy	White to pink	21.54	3-4	23.38×2.45	0	7.52×2.31
17	Fol 17	Ponchamalli	Fluffy	White	26.90	3-4	26.65×2.18	0	8.43×2.67
18	Fol 18	Krishnapuram	Fluffy	Pink	22.42	2-3	24.35×2.57	0	7.94×2.52
19	Fol 19	Machery	Sparse	Grey	20.44	2-3	22.57×2.45	0	7.52×2.31
20	Fol 20	Penagram	Sparse	White	18.79	2-3	22.16×2.45	0	7.32×2.14
		CD (0.05)					**2.04**		

Table 3 Virulence of different F. oxysporum f. sp. lycopersici isolates

Sl. No.	District	Place of collection	Isolate	Soil inoculation method		
				DAI	Wilt %	Pathogenecity
1	Tirunelveli	Sivagiri	Fol1	30	100	Highly virulent
2	Krishnagiri	Ponchamalli	Fol17	30	96	Highly Virulent
3	Tirunelveli	Athuvazhi	Fol2	30	78	Virulent
4	Krishnagiri	Krishnapuram	Fol18	35	84	Virulent
5	Salem	Athur	Fol15	35	72	Moderate
6	Ariyalur	Veerakkan	Fol5	35	70	Moderate
7	Madurai	Solavanthan	Fol3	35	57	Less virulent
8	Coimbatore	Thondamuthur	Fol14	35	55	Less virulent
9	Ariyalur	Pillakurichi	Fol6	35	53	Less virulent
10	Virudhunagar	Sethur	Fol11	35	52	Less virulent

cent on the 30th day, whereas Fol. 6 and Fol. 11 recorded the least PDI of 53 and 52Per cent on the 35th day of inoculation respectively. Similar results have been reported by Alice, (1994). The results indicated that difference in morphological character positively correlated with its virulence.

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