Spatial distribution, isolation, and characterization of morphological variants of the *Fusarium* Wilt pathogen in brinjal across Andhra Pradesh and Tamil Nadu, India

ABSTRACT

Fusarium wilt, caused by various *Fusarium* species, is a devastating disease of brinjal (*Solanummelongena*) leading to substantial yield losses in major production regions. This study aimed to assess the prevalence of *Fusarium* wilt in Tamil Nadu and Andhra Pradesh during 2024. A comprehensive survey across brinjal-growing regions, including Chittoor, Kurnool, Guntur, Salem, Dindigul, Krishnagiri, and Cuddalore, revealed varying disease incidence. Hosur (Tamil Nadu) exhibited the highest percent disease incidence (PDI) at 56.45%, followed by Madanapalli (Andhra Pradesh) at 54.67%. Pathogen isolation and identification using the tissue segment method confirmed *Fusarium* sp. as the primary causal agent. Morphological characterization revealed significant variations among isolates in colony shape, conidial diameters, and growth patterns, indicating a heterogeneous *Fusarium* population across the investigated locations. To lessen the effect of *Fusarium* wilt on brinjal output, these results highlight the vital necessity for ongoing monitoring, precise pathogen identification, and the creation of integrated management techniques such resistant variety breeding.

Keywords:Solanummelongena, *Fusarium*, Percent Disease Incidence (PDI), Pathogen identification, Morphological characterization, Heterogeneity, Integrated management.

INTRODUCTION

Vegetables are essential to the human diet because they offer a variety of nutrients. A regular diet that includes fresh vegetables in a balanced manner can lower the chance of developing chronic illnesses (Joshipura et al. 2001). According to Kumar et al. (2015), solanaceous vegetables are protective foods because they are high in important vitamins, minerals, dietary fibers, and phytonutrients. There are over 3,000 species in the Solanaceae family, which includes many herbs, medicinal plants, and industrial crops in addition to some of the most often consumed vegetables, including tomato, eggplant, chillies, and peppers (Vorontsova and Knapp 2012). Brinda and aubergine are other names for eggplant (*S. melongena*.), which is one of the five most significant vegetables produced worldwide (FAO 2021).Berries are the fruit of this perennial, non-tuberous crop. Eggplant is mostly seeded in late spring and needs a long, warm growing season due to its 100–120 day growth cycle.In eggplant, secondary metabolites including phenolic chemicals help control diabetes, decrease cholesterol, and lessen the risk of heart disease (Serdula et al. 1996; Jenkins et al. 2003). According to reports, eggplant contains three main types of phenolics: polyphenols, flavonoids, and phenolic acids (Stommel and Whitaker 2003; Sharma and Kaushik 2021). In order to regulate glucose absorption and to manage type 2 diabetes, phenol-enriched eggplant extracts show significant inhibition of α-glucosidase and angiotensin-converting enzymes (Kwon, Apostolidis, and Shetty 2008). Unfastened antioxidants that scavenge radicals are abundant in eggplant (Luthria 2006). Eggplant is a vegetable of choice for a healthy lifestyle due to its high antioxidant and phenolic contents (Urquiaga and Leighton 2000; Sekara, Cebula, and Kunicki 2007). Eggplant originated in India (Zeven and Zhukovsky 1975). With 57% and 27% of the global production,

respectively, China and India are the two largest producers of eggplant (Frary, Doganlar, and Daunay 2007). In other Asian nations including Bangladesh, Pakistan, and the Philippines, the crop is also widely farmed in their warm climates. 13.154,000 metric tons of eggplant were produced on 758,000 hectares of land in 2020–2021, according to the Ministry of Agriculture and Farmers Welfare, Government of India (Ministry of Agriculture and Farmers Welfare, Government of India 2020–2021). Both human interventions, including as planting, propagation, reselection, and advanced cropbreeding programs, and the ongoing natural selection process have produced the eggplant varieties that are now grown (Ramesh and Vasanthi 2008).Consumer preferences, such as flavor, color, and cooking qualities, which fluctuate significantly even throughout Indian states, were the primary motivators for the human interventions .The signs of fusarium wilt include the plant's yellowing and withering, as well as the leaves rolling inward and upward. The plant may wilt and eventually die in a matter of days as a result of the younger leaves dying one after the other (Sankara Reddy and Rajamohan, 2022). Due to its high frequency and severity, Fusarium wilt requires an integrated strategy to disease control that combines biological, resistant cultivars, accurate diagnostic technologies, and effective cultural methods.

Materials and Methods

Survey and collection of Fusarium spp. isolates from different regions of Andhra Pradesh and Tamil Nadu

Major districts that cultivate brinjal in Tamil Nadu (Salem, Krishnagiri, and Dindigul) and Andhra Pradesh (Chittoor, Kurnool, and Guntur) were surveyed in 2024(Table 1). In order to determine the occurrence of Fusarium wilt in various regions of Andhra Pradesh and Tamil Nadu, samples are taken at the crop's maturity development stage. Based on the diagnosis of common Fusarium wilt disease signs, including leaf yellowing, wilting, vascular browning, and plant mortality, infected plants were found. Diseased samples are gathered by choosing several plots from each village and calculating the disease incidence. Random sampling has been carried out along the roadside at intervals of 5–10 km and in some inner villages. This method was used to get the average percent disease incidence (PDI).

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

Isolation and identification and purification of the pathogen

The tissue segment technique (Agrios, 2005) was used to isolate the wilt pathogenic isolates from the infected root and stem samples after they were cultivated in PDA medium. Samples of infected roots and stems were blotted after being rinsed for ten minutes under running water. (Katan, 1971),

Brown vascular bundles were chosen for isolation since the infection is restricted to these. After being cut into small pieces (3-5 mm) and surface sterilized with 1% sodium hypochlorite (NaOCl) for 30 seconds, the infected vascular bundles and healthy portions were rinsed three times in various sterile distilled water variations (Dongzhen et al., 2020) and dried with tissue paper. To avoid bacterial contamination, two to four pieces were added to a Petri plate with solidified potato dextrose agar (PDA) medium supplemented with streptomycin sulphate. The culture was then cultured for three to five days at 28±2 °C. The culture was kept in agar slants at 25±2 °C after being purified using the single spore isolation technique (Leslie and Summerell, 2008).

Morphological features of Fusarium sp.

Based on the morphology of the colonies, the characteristics of the macroconidia and microconidia, and the measurement of chlamydospores, twenty isolates of Fusarium were identified (Table 2). The Fusarium isolates were identified by matching their morphology to the descriptions found in the Fusarium atlas (Leslie and Summerell, 2006; and Samson et al. 2008) and reference manual (Booth, 1971).The growth and morphological characteristics of the isolates—colony morphology, mycelial growth, colony color, and conidia size, shape, and septation—are used to identify the pathogen (Soesanto et al., 2011). An LR-HD trinocular microscope with a 5MP HDMI camera was used to measure the sizes of macroconidia, microconidia, and chlamydospores. At 400X magnification, phase contrast pictures of the macroconidia and microconidia were taken (Tsegaye and Tesfaye, 2020).

Results and Discussion

Survey on the incidence of Fusarium wilt of Brinjal from major chilli growing areas of Andhra Pradesh and Tamil Nadu

An intensive survey of major Brinjal growing areas in Andhra Pradesh and Tamil Nadu reveals that the disease is endemic.Hosur (F17) had the highest diseases incidence of any of the places studied, with 56.45. Madanapalli (F2), Thalaivasal (F11), Anaipatti (F14), Velugodu (F4), and Parathurchavadi (F20) had the next highest rates, with 54.67, 50.54, 48.12, 46.19, and 41.46, respectively.The disease's incidence was moderate in Tenali (39.37), KK Nagar (37.98), Ponnur (35.59), and Kuppam (32.54), while Sundekuppam (10.12) had the lowest wilt incidence. The incidence of the condition was significantly greater in Tamil Nadu than in Andhra Pradesh, according to the report. The disease susceptibility of the kinds cultivated in various soil types and places may be the cause.The vulnerability of Pusa Purple Long types in comparison to local varieties produced across the areas is also revealed by the earlier data from the other surveyors; all of the data is included in Table 1. across their survey, Birla (2014) evaluated the prevalence of Fusarium.sp. caused chilli wilt across the Nimar Valley and Malura Plateau zone. They found that Khargone had the highest average disease incidence, at 34.93%. According to Manasa et al. (2022), who carried out a roaming study in tomato-growing regions of Andhra Pradesh's Rayalaseema zone in 2021, the disease prevalence ranged from 15 to 60%. Chinnahuithy village in Kurnool district had the highest PDI (59.5%), followed by Settivaripalle in YSR Kadapa (48%), while Anantharajupeta in YSR Kadapa had the lowest PDI (15%). Geographical locations

and the isolates' varying levels of virulence may be the cause of the observed variances in disease incidence.In all of India's chilli-growing regions, Fusarium wilt has recently become a more serious disease (Singh et al., 1998; Nishani et al., 2021). In Karnataka, it can cause yield losses of up to 25% (Madhukar and Naik, 2004; Mishra et al., 2018), 0.0–70.0 (Ravikumara et al., 2022), and 15–25% in arid regions of Pakistan (Siddiqui et al., 2007).Wilt incidence ranged between 10 and 80 percent (Devika Rani, 2006) and 0.0 to 75.0 percent (Anonymous, 2005). The average recovery percentage of Fusariumoxysporum sp.-caused Fusarium wilt in several regions of India was 32.54 percent.

Identification and morphological characters of the pathogen

On potato dextrose agar medium, all Fusarium spp. isolates develop white, creamy, pale brown, brownish-colored fluffy, and cottony mycelial growth. Isolates F17 and F2 from Hosur and Madanapalli had the highest mycelial growth rates of 90.00¬ and 89.56. After 10 days of plating, Thalaivasal (F11), Anaipatti (F14), Velugodu (F4), and Parathurchavadi (F20) received 88.19, 87.16, 85.49, and 83.94, respectively. The isolates F7, F16, F9, and F1 are decreasing, with isolate F18 from Sundekuppam village in Krishnagiri district, Tamil Nadu, having the lowest mycelial growth, as shown in Table 2 (Figure 2). The production of microconidia, macroconidia, and chlamydospores by 20 isolates is documented and shown in table 2. The size of microconidia varies from 12.67-25.91 µm length to 3.18- 7.35 µm breath, while macroconidia ranges from 38.68-59.97 µm length to 5.45-8.67 µm. The diameter of chlamydospores ranges from 9.72-11.85 µm, with 0-1 and 2-5 septa, respectively (Table 2). All conidia are sickle to crescent shaped with blunt edges, and chlamydospores are terminal to intercalary, round to oval in form (Figure 3).All twenty Fusarium sp. isolates cultured on potato dextrose agar (PDA) medium exhibited a variety of growth patterns and colony characteristics, ranging from white, fluffy to cream white cottony mycelium. The isolates F1, F4, F5, F10, and F19 yield creamy white cottony mycelium, while F8, F9, F11, F13, and F17 produce pale brown to white mycelium (Table 1). The pathogen is recognized using the morphological and cultural characteristics provided by Butler, 1910; Padwick, 1940; Booth 1971; and Leslie and Summerell (2006). Oljira and Berta (2020) isolated and characterized the Fusarium wilt of pepper pathogen from Ethiopia's Gurgeon zone, identifying Fusariumoxysporum f. sp. capsici as the cause of the illness. Similarly, Hami et al. (2021) observed that F. equiseti was responsible for chlli wilt in the Kashmir area. Additionally, reports of F. oxysporum and F. solani have been made from several regions of India, including the Kashmir Valley (Rajeswari and Kannabiran, 2011) and Naik et al. (2008). The findings are consistent with those of Soleha et al. (2022), who determined the cause of the Fusariumoxysporum-induced acacia seedling wilt disease in south Sumatra.

Table 1. Survey and Isolation of *Fusarium*Spp infected plants from different districts of Tamil Nadu & Andhra Pradesh.

S.No.	Isolate	Area	Districts	Variety	Soil type	Coordinates	Fusarium Wilt incidence
1	F1	Chittoor	Kuppam	Swarna	Red soil	12°44'53"N 78°19'39"E	32.45^{h} (34.72)
2	F2		Madanapalli	Nidhi	Red soil	13°33'58"N 78°28'58"E	54.67^{a} (48.71)
3	F3		Ramakuppam	Swarna	Red soil	12°53'42"N 78°28'32"E	21.12^{m} (27.70)
4	F4	Kurnool	Velugodu	Nidhi	Red soil	15°42'56"N 78°34'36"E	46.19^{d} (43.25)
5	F5		Peddapadu	Swarna	Black soil	15°48'39"N 77°59'11"E	30.79^{i} (33.83)
6	F6		Atmakur	Swarna	Red soil	15°52'31"N 78°34'06"E	20.45^{m} (26.88)
7	F7	Guntur	Tenali	Nidhi	Red soil	16°14'32"N 80°40'00"E	39.37^{f} (38.85)
8	F8		Rompicharala	Anand	Black soil	16°12'23"N 79°54'49"E	18.37^{n} (25.37)
9	F9		Ponnur	Anand	Black soil	16°04'15"N 80°34'04"E	35.59^{g} (36.62)
10	F10	salem	Attur	Pusa Purple Long	Black soil	11°36'33"N 78°35'33"E	24.91^{l} (29.88)
11	F11		Thalaivasal	Pant Bahar	Black soil	11°35'11"N 78°45'45"E	50.45^{b} (45.25)
12	F12		Gangavalli	Pant Bahar	Red soil	11°30'02"N 78°39'00"E	28.32^{j} (32.27)
13	F13	Dindigul	Seelapadi	Annamalai	Black soil	10°24'05"N 77°59'43"E	15.45^{op} (23.42)
14	F14		Anaipatti	Pusa Purple Long	Red soil	10°23'57"N 77°54'54"E	48.12^{c} (44.38)
15	F15		Siluvathur	Pant Bahar	Red soil	10°21'53"N 78°04'51"E	12.61^{q} (20.87)
16	F16	Krishnagiri	KK Nagar	Pusa Purple Long	Red soil	12°31'27"N 78°16'41"E	37.98^{f} (38.04)
17	F17		Hosur	Pusa Purple Long	Red soil	12°46'44"N 77°49'46"E	56.45^{a} (48.71)
18	F18		Sundekuppam	Pant Bahar	Black soil	12°27'10"N 78°13'11"E	10.21^{r} (18.6)
19	F19	Cuddalore	Sivapuri	Annamalai	Black soil	11°21'30"N 79°43'14"E	26.34^{k} (30.87)
20	F20		Parathurchavadi	Annamalai	Black soil	11°25'37"N 79°34'32"E	32.45^{h} (34.72)

*Mean of three replications

*Values in the column followed by common letters do not differ significantly at 5% level by Duncan's multiple range test (DMRT)

Isolates	Colony Characters	Mycelial growth (mm) 10 DAI	Micro conidia			Macro conidia			Chlamydospore
			Length (µ)	Width (µm)	No. of Septation	Length (µ)	Width (µm)	No. of Septation	Diameter
F1	Cream white mycelium	74.25^{fg} (59.50)	18.75	3.67	0	44.05	7.50	3-5	11.85
F2	White fluffy cottony mycelium	89.56^{ab} (71.76)	12.67	5.15	0-1	46.56	6.30	2-4	10.55
F3	White cottony mycelium	62.49^{ij} (53.10)	17.13	3.56	0-1	56.50	6.03	2-3	10.00
F4	Cream white submerged cotton mycelium	85.49^{bc} (68.82)	25.91	6.51	0	41.65	5.98	3-5	11.5
F5	Cream white submerged cotton mycelium	71.58^{fg} (58.08)	15.25	4.67	0	45.86	7.56	5-6	10.05
F6	White fluffy cottony mycelium	61.82^{ij} (51.84)	23.21	7.35	0	57.74	7.07	3-5	12.01
F7	White fluffy raised cottony mycelium	80.26^{de} (63.71)	21.98	5.84	0-1	65.40	5.45	3-5	9.95
F8	Pale brown to white cottony mycelium	59.71^{jk} (50.60)	18.34	3.63	0	59.97	8.67	2-4	10.8
F9	Pale brown to white cottony mycelium	76.34^{ef} (60.93)	17.25	4.59	0-1	53.12	7.45	2-4	9.95
F10	Cream white submerged cotton mycelium	64.89^{ij} (53.53)	24.87	6.15	0-1	55.46	6.82	3-5	10.5
F11	Pale brown to dark brown zonation	88.19^{ab} (69.90)	16.67	4.09	0	54.62	8.05	2-3	10.4
F12	White cottony mycelium	69.12^{gh}(56.53)	17.54	4.26	0-1	40.91	6.29	3-4	10.36
F13	Pale brown to dark brown zonation	58.46^{jk} (50.66)	16.67	3.18	0	44.78	6.06	3-5	11.02
F14	White cottony mycelium	87.16^{ab} (70.31)	15.56	3.76	0	55.56	6.67	2-3	9.80
F15	Brownish mycelium	55.24^{kl} (48.22)	20.87	4.69	0-1	39.84	5.51	2-3	10.18
F16	Milkfish white cottony mycelium	79.92^{de} (63.39)	15.26	3.45	0-1	38.68	5.57	2-4	9.85

F17	Pale brown to white cottony mycelium	90.00[a] (71.98)	18.54	4.91	0-1	42.76	6.65	2-4	9.72
F18	Milkfish white cottony mycelium	54.16 [l](47.38)	18.34	4.12	0	43.10	5.77	3-4	10.58
F19	Creamy white fluffy mycelium	66.46[hi] (54.62)	22.85	6.32	0	48.23	6.81	3-4	9.80
F20	Brownish mycelium	83.94[cd] (66.18)	17.52	3.98	0-1	45.56	5.98	2-4	9.45

Table 2. Morphological and cultural characters variability of *Fusarium* Sp. From different Localities of Tamil Nadu & Andhra Pradesh.

Fig 1 : Axenic culture of Fusariumincarnatum**Fig 2:** Mycelial growth of twenty *Fusarium* sp. isolates.

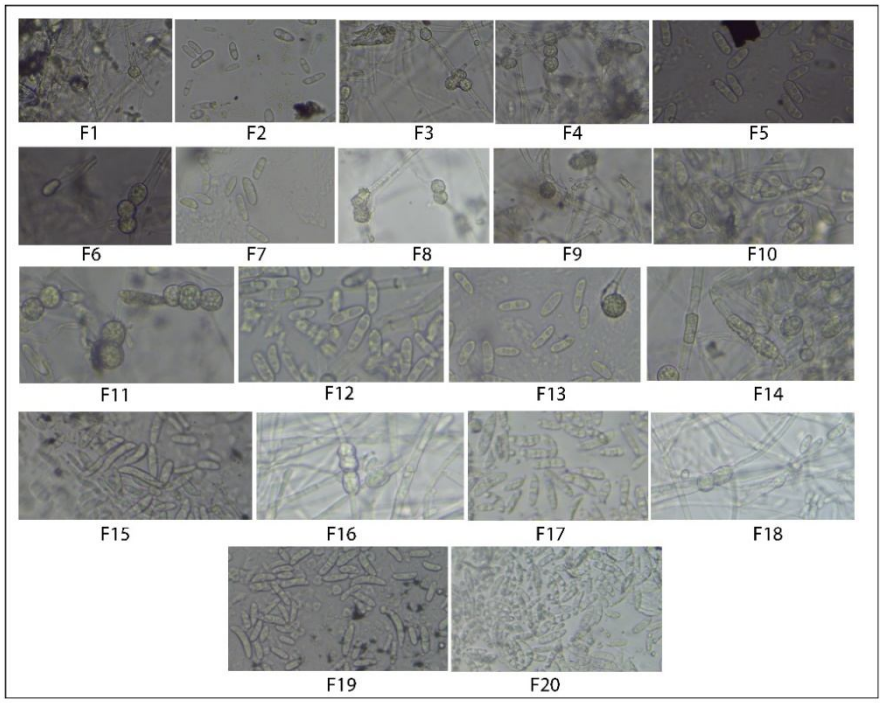

Fig 3: A microscopic image of conidia from 20 distinct Fusarium sp. isolates.

Conclusion

This study provides a comprehensive assessment of the incidence of Fusarium wilt in brinjal crops in Tamil Nadu and Andhra Pradesh's primary growing regions. According to the paper, fusarium wilt is a frequent and deadly disease that affects brinjal farming, with severity varying by place. The highest sickness incidence was discovered in Hosur (F17) in Tamil Nadu followed by Madanapalli (F2) in Andhra Pradesh and other locations. The changes in disease incidence may be attributed to differences in isolate virulence, soil characteristics, and cultivar susceptibility. Fusarium spp. were isolated and identified using tissue segmentation methods, and morphological analysis confirmed the presence of several Fusarium species. The isolates' conidia characteristics, mycelial growth, and colony morphology differed. This study underlines the importance of ongoing monitoring and identification of Fusarium species in order to develop effective management strategies. The findings indicate the need for more research on resistant cultivars and integrated disease management measures in chilli crops to decrease production losses caused by Fusarium wilt.

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