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Survey and Characterization of Fusarium oxysporum f. sp. cubense Associated with Fusarium Wilt of Banana in Major Growing Regions of Tamil Nadu, India

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

The survey-based experimental study was conducted from July 2023 to May 2024 at the Department of Plant Pathology, Faculty of Agriculture, Annamalai University, Tamil Nadu, and in banana fields across major banana-growing regions of the state. Surveys were carried out in twenty locations covering six districts, representing AAA, AAB, ABB, and AB genome groups under different soil types. Infected pseudostem tissues were surface-sterilized and cultured on potato

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dextrose agar medium, and the resulting isolates were characterized based on colony morphology and conidial dimensions of macroconidia, microconidia, and chlamydospores. Genomic DNA was extracted using the CTAB method, and the ITS region was amplified with universal primers ITS4/5, sequenced, and analyzed through BLAST and phylogenetic inference in MEGA 7. A total of twenty morphologically distinct isolates were obtained, and their cultural and conidial features were documented. The BLAST search confirmed the identity of the isolates as *Fusarium oxysporum* and *Fusarium oxysporum* f. sp. *cubense* with an amplicon size of 500 bp. The study revealed that banana cultivation in Tamil Nadu was severely affected by *Fusarium oxysporum* f. sp. *cubense*, thereby highlighting the urgent need for region-specific disease management strategies, particularly the identification and promotion of resistant cultivars. Furthermore, continuous molecular surveillance and pathogen monitoring were considered essential for detecting the emergence of highly virulent strains and for ensuring sustainable banana production in the region.

Keywords: Fusarium wilt; morphological characterization; pathogenicity; molecular identification.

1. INTRODUCTION

Banana (Musa spp.) is a vital fruit crop cultivated extensively in tropical and subtropical regions owing to its significant economic and nutritional value on a global scale (Chukwu et al., 2025; Maseko et al., 2024). Belonging to the family Musaceae, it is believed to have originated in the Indo-Malayan region of Southeast Asia. Commonly referred to as the "fruit of the wise", banana ranks as the fourth most important staple food worldwide (Pareek, 2016). It serves as an excellent source of potassium and provides high levels of carbohydrates, essential vitamins particularly Bcomplex vitamins and minerals, making it a key component of the human diet (Vieira et al., 2025). The fruit is easily digestible and naturally free from fat and cholesterol (Kumari et al., 2023; Ranjha et al., 2022). In India, Tamil Nadu stands as the fourth largest producer of bananas, contributing approximately 3,895.64 thousand metric tonnes, accounts for 10.41% of the national output. During the 2022–2023 cultivation period, the state reported a production of 4,522.6 thousand metric tonnes with an average productivity of 41.2 metric tonnes per hectare, across an area of 109.7 thousand hectares (Vignesh et al., 2023).

However, banana cultivation is severely challenged by various abiotic and biotic stresses. Abiotic factors include soil moisture deficit, salinity, extreme temperatures, and strong winds (Abdoussalami et al., 2023; Dhanyasree et al., 2022). Major biotic constraints include fungal, bacterial, and viral diseases such as Sigatoka leaf spot, Fusarium wilt, Xanthomonas wilt, Moko disease, freckle leaf spot, banana blood disease, and banana

bunchy top virus (Esquivel Chávez et al., 2025; Tinzaara et al., 2024). Among these, Fusarium wilt caused by Fusarium oxysporum f. sp. cubense (Foc) is regarded as one of the most destructive diseases, significantly impacting banana yield worldwide (Ismaila et al., 2023).

Fusarium wilt initially presents internal symptoms, notably reddish-brown discoloration of the xylem in the feeder roots. This is subsequently followed by external symptoms, beginning with chlorosis in older leaves, which progresses from the leaf margins toward the midrib. As the disease advances, petioles darken and collapse, and longitudinal cracks develop along the pseudostem. Senescing leaves droop and form a distinctive skirt-like arrangement around the base of the plant. In the later stages of infection, the heart leaf wilts while the pseudostem remains erect. Internally, a purple to brown discoloration is prominent within the vascular tissues, particularly at the stele-cortex junction. The pathogen is capable of systemic movement through young suckers, facilitating its spread within plantations. Infected plants often exhibit poor fruit development, producing small, fibrous and acidic fruits or in some cases, no fruit at all. Foc survives and propagates through the production of macroconidia, microconidia and thick-walled chlamydospores, which contribute to its persistence in soil environments (Costa et al., 2025; Sasaki et al., 2025; Thangavelu et al., 2020).

This pathogen is highly adaptable, thriving in a wide range of soil pH conditions from 4.8 to 8.4and across various soil textures, including sandy loam and heavy clay. Optimal growth of Foc occurs at approximately 28°C, while temperatures exceeding 33°C or falling

below 17°C inhibit its development (Dita et al., 2018). The present study aims to investigate the morphological and molecular characteristics of *Fusarium oxysporum* f. sp. *cubense*, the causal agent of Fusarium wilt in banana, to support accurate identification and inform effective disease management strategies.

2. MATERIAL AND METHODS

2.1 Field Survey of Fusarium Wilt in Banana Across Major Regions of Tamil Nadu

A roving survey was carried out across 20 farms located in six major banana-growing districts of Tamil Nadu, India, during the period 2023-2024. The objective was evaluate the incidence of Fusarium wilt in banana and to collect samples of Fusarium oxysporum f. sp. cubense (Foc) for subsequent isolation and characterization. The survey included major banana cultivars commonly grown in the respective regions. At each surveyed site, a minimum of 50 to 100 plants were systematically examined for typical Fusarium wilt symptoms. From each farm, two to representative samples exhibiting characteristic disease symptoms were collected for laboratory analysis and pathogen isolation (Thangavelu et al., 2024).

2.2 Collection of Samples

Symptomatic banana plants exhibiting typical Fusarium wilt indications, including reddishbrown discoloration of the rhizome pseudostem, were sampled from twentv locations across six major banana-growing districts of Tamil Nadu. From each affected plant, discoloured vascular tissues were aseptically excised, wrapped in sterile paper towels and sealed in properly labelled envelopes with details of the sampling site and cultivar (Thangavelu et al., 2024). The collected specimens were subsequently transported to the Department of Faculty Pathology, of Agriculture, Annamalai University, India, for further analysis.

2.3 Isolation of Pathogen

In the laboratory, vascular strands exhibiting discoloration were cut into 5–8 mm fragments and surface-sterilized by sequential immersion in

1% sodium hypochlorite followed by 70% ethanol, each for 30 seconds. The treated tissues were then rinsed thoroughly five times with sterile distilled water and air-dried on sterile blotting sheets. From each sample, four dried fragments were aseptically placed onto quarterstrength dextrose potato agar supplemented with 0.5% streptomycin sulfate to inhibit bacterial contamination and incubated at 25 °C for four days. Emerging fungal colonies were subsequently sub-cultured and purified using the hyphal tip method. The purified Fusarium isolates were preserved in 15% glycerol at -80 °C for long-term storage and maintained on quarter-strength PDA slants at 4 °C for subsequent studies (Thangavelu et al., 2024).

2.4 Morphological Identification of Fusarium oxysporum Isolates

Fungal isolates were characterized based on their colony appearance and morphological features. The cultures were incubated at 28 ± 1 °C for seven days, during which cultural characteristics were periodically recorded. For microscopic assessment of spore morphology, seven-day-old cultures grown on quarter-strength potato dextrose agar (PDA) were examined Zeiss AX10 stereomicroscope а (Göttingen, Germany) fitted with an Art Cam 300 MI digital camera (Artray, Tokyo, Japan). The emphasized observations the structural characteristics of microconidia, macroconidia and chlamydospores (Cha et al., 2007).

2.5 Pathogenicity Test

2.5.1 Inoculation

Three-month-old, disease-free tissue-cultured banana plantlets of the cultivar Ney Poovan (AB group) and transplanted into 10 x 15 cm grow bags. Each grow bag was filled with 7 kg of a sterilized mixture of red soil, sand, and farmyard manure in a 1:1:1 (w/w/w) ratio. The *Fusarium* inoculum was prepared by incubating 5–6 agar plugs (9 mm diameter) of actively growing fungal culture in a sterile substrate composed of 900 g sand, 100 g maize meal, and 200 mL distilled water. The substrate was autoclaved twice and incubated at 25°C for 15 days, with thorough mixing at 5-day intervals to ensure uniform fungal colonization (Thangavelu et al., 2024).

Table 1. Standardized Scoring System for Evaluating External and Internal Symptoms of Fusarium Wilt in Banana under Greenhouse Conditions

Score	External Symptoms Description	Internal Symptoms description
1	No symptoms	No symptoms
2	Initial yellowing mainly in the lower leaves	Initial rhizome discolouration
3	Yellowing of all the lower leaves with some discolouration of younger leaves	Slight rhizome discolouration along the whole vascular system
4	All leaves with intense yellowing	Rhizome with most of the internal tissues showing necrosis
5	Plant dead	Rhizome fully necrotic

2.5.2 Virulence in pot culture

A total of twenty Fusarium isolates obtained from diseased banana plants were subjected to pathogenicity assays. For each isolate, 30 g of colonized substrate was incorporated 2-3 cm below the soil surface at the base of individual plants. The experiment was arranged in a completely randomized design with three biological replicates per isolate, while noninoculated plantlets served as negative controls (Thangavelu et al., 2019). All plants were maintained under greenhouse conditions, and disease assessment was carried out 30 days post-inoculation. Symptom severity was rated on a 1-5 scale described by (Pérez-Vicente & Dita, 2014), based on both external and internal manifestations. Fungal re-isolation symptomatic pseudostem tissues confirmed the pathogenicity of the isolates. After incubation at 28 ± 1 °C for 3–5 days, the isolates were preserved in 50% glycerol at -80 °C for longterm maintenance.

2.6 Molecular Identification of Fusarium oxysporum Isolates

Genomic DNA was isolated from the fungal cultures usina modified а cetyltrimethylammonium bromide (CTAB) method. The internal transcribed spacer (ITS) region was amplified with the universal primer pair ITS4 and ITS5. PCR amplification was carried out with an initial denaturation at 94 °C for 10 minutes, followed by 35 cycles of denaturation at 94 °C for 30 seconds, annealing at 54 °C for 30 seconds, and extension at 72 °C for 30 seconds, with a final extension at 72 °C for 10 minutes. The amplified products were sequenced by Biofocus Scientific Solutions Pvt. Ltd. (Kumbakonam, India), and the obtained sequences were subjected to BLAST analysis against the NCBI nucleotide database for identification. Based on maximum identity scores, the top ten sequences were aligned using Clustal W for multiple sequence alignment. A distance matrix was generated, and a phylogenetic tree was constructed using MEGA 7 software. The validated sequences subsequently deposited in the NCBI GenBank database and assigned accession numbers. For species-level identification. the translation elongation factor 1-alpha (TEF-1α) gene was amplified using specific primers. In addition, race-specific detection was carried out with primers **FOC** R1 F'(TACCTCCTTGGTCGACAGGT) and R'(CAGACTTCCAACGTCTCGGT) and FOC R4 F'(CGCACTCTTACGTTGAGGAT) (TCCACGCAACACTAGCTACT) to differentiate between the respective lineages (Thangavelu et al., 2024).

3. RESULT AND DISCUSSION

3.1 Field Survey and Disease Incidence Analysis of Fusarium Wilt in Banana Across Major Regions of Tamil Nadu

A field survey conducted at 20 distinct locations of six district in Tamil Nadu assessed the incidence of Fusarium wilt in banana cultivars The during 2023-2024. percent disease incidence (PDI) showed notable variation among isolates, cultivars, and geographic regions (Table 2). The highest PDI (69.43%) was observed in the Nev Poovan cultivar (AB genome) from Kokilapuram, followed by 63.63% in the Grand Nine cultivar (AAA genome) from Anaipatti. In contrast, the lowest incidence (9.28%) was recorded in the Karpooravalli cultivar (ABB genome) from Madukkarai.

Geographical variation in PDI across surveyed locations may be attributed to differences in soil conditions, microclimate, agronomic practices, and the presence of diverse Foc races or strains. Similar findings have been documented in previous studies conducted in other banana-

Table 2. Survey on the incidence of Banana Fusarium wilt in major banana growing areas

S. No	Isolate	Village	Cultivar	Crop Stage (Months)	Genome	Percent Disease Incidence (%)	Average disease Incidence (%)
1	AUFO1	Chinnaovulapuram	Grand nine	7	AAA	51.09	55.60 (48.22)
2	AUFO2	Nayaranathevan Patti	Grand nine	8	AAA	58.56	
3	AUFO3	Lower camp	Grand nine	8	AAA	33.28	
4	AUFO4	Anaipatti	Grand nine	7	AAA	63.63	
5	AUFO5	Kokilapuram	Ney Poovan	9	AB	69.43	
6	AUFO6	Pinnathur	Monthan	8	ABB	16.36	14.06 (22.02)
7	AUFO7	Ayepettai	Monthan	9	ABB	11.76	
8	AUFO8	Thottiyam	Ney Poovan	9	AB	45.25	41.73 (40.24)
9	AUFO9	Lalgudi	Ney Poovan	7	AB	35.19	,
10	AUFO10	Manachanallur	Ney Poovan	8	AB	38.92	
11	AUFO11	Kannanur	Ney Poovan	9	AB	47.54	
12	AUFO12	Gobichetti palayam	Grand nine	9	AAA	32.61	30.51 (33.53)
13	AUFO13	T.N. Palayam	Grand nine	7	AAA	40.64	
14	AUFO14	Anthiyur	Ney Poovan	8	AB	34.73	
15	AUFO15	Arasalur	Ney Poovan	9	AB	30.08	
16	AUFO16	Thondamuthur	Karpooravalli	8	ABB	17.06	12.63 (20.82)
17	AUFO17	Madukkarai	Karpooravalli	8	ABB	9.28	,
18	AUFO18	Karamadai	Karpooravalli	7	ABB	11.54	
19	AUFO19	Mohanur	Rasthali	8	AAB	23.21	23.07 (28.71)
20	AUFO20	Neikkaran patti	Rasthali	7	AAB	22.94	` '

growing regions, which emphasize the role of both biotic and abiotic factors in influencing disease expression (Olivares et al., 2021; Thangavelu et al., 2024).

3.2 Morphological Characterization of Fusarium oxysporum Isolates

Twenty isolates of Fusarium oxysporum were characterized based on their colony morphology on potato dextrose agar (PDA) after 7 days of incubation at 28 ± 1°C. The isolates displayed variation in colony texture, considerable pigmentation, margin regularity, and shape. Colony types ranged from profuse fluffy to flat humid and cottony textures, with pigmentation varying from white and pale yellow to pinkish and purple tones (Table 3 and Fig. 1). Most colonies exhibited smooth margins and circular forms, while some were irregular. Radial growth after 7 days ranged between 6.21 cm and 9.00 cm, indicating differential growth dynamics among the isolates. The isolates exhibited a wide range of colony characteristics, including differences in texture (fluffy, cottony, flat humid). or

pigmentation (ranging from white to pink and purple), and colony margin morphology.



Fig. 1. Pure culture of *Fusarium oxysporum* f.sp. *cubense*

These phenotypic differences are consistent with earlier reports highlighting the polymorphic nature of *F. oxysporum*, which often varies with substrate, isolate origin, and environmental conditions such as temperature and incubation time (Garzón-Nivia et al., 2025; Harish et al., 2023).

Table 3. Cultural characteristics of Fusarium oxysporum

S. No	Isolate	Colony character	Pigmentation	Margin	Shape	Radial growth @ 7days (cm)
1	AUFO1	Profuse fluffy	White	Smooth	Circular	8.49
2	AUFO2	Less fluffy	White	Smooth	Irregular	8.12
3	AUFO3	White with	Purple	Smooth	Circular	8.23
		concentric rings	·			
4	AUFO4	White fluffy	Pinkish	Smooth	Irregular	8.55
5	AUFO5	White cottony	White with purple	Smooth	Circular	9.00
			centre			
6	AUFO6	Moderately fluffy	White	Smooth	Circular	8.42
7	AUFO7	White fluffy	Slightly purple	Smooth	Irregular	9.00
8	AUFO8	White cottony	Pale pink	Smooth	Circular	9.00
9	AUFO9	Flat humid	White	Irregular	Circular	7.92
10	AUFO10	Profuse fluffy	Pale yellow	Smooth	Circular	8.69
11	AUFO11	Raised fluffy	White	Smooth	Circular	8.68
12	AUFO12	White cottony	White with purple	Smooth	Circular	9.00
			centre			
13	AUFO13	White thick	White	Irregular	Irregular	8.64
14	AUFO14	Abundant	Pale yellow	Smooth	Circular	7.65
15	AUFO15	White cottony	White	Smooth	Circular	8.56
16	AUFO16	White with	Slightly purple	Smooth	Irregular	6.21
		concentric rings				
17	AUFO17	Flat humid	White	Irregular	Circular	6.73
18	AUFO18	White cottony	White with slightly	Smooth	Irregular	7.00
			purple			
19	AUFO19	Flat humid	White	Smooth	Irregular	8.18
20	AUFO20	Flat humid	White	Irregular	Circular	7.40

Table 4. Conidial characteristics of Fusarium oxysporum

S. No	Isolate	Macroconidia (μm)			Microconidia (μm)			Chlamydospore	
		Length	Breadth	Shape	Length	Breadth	Shape	Size (µm)	ShapeSS
1	AUFO1	15.01	2.63	Fusiform	4.87	1.02	Oval	7.63	Oval
2	AUFO2	17.63	2.47	Cylindrical	5.61	2.45	Oval	7.12	Oval
3	AUFO3	21.03	2.68	Fusiform	5.02	1.09	Cylindrical	7.95	Oval
4	AUFO4	14.23	2.98	Fusiform	4.98	2.47	Cylindrical	8.41	Round
5	AUFO5	16.36	4.47	Fusiform	4.80	2.64	Oval	8.03	Oval
6	AUFO6	15.31	2.78	Fusiform	5.96	2.61	Fusiform	7.19	Oval
7	AUFO7	13.28	3.29	Fusiform	6.36	2.70	Oval	8.63	Round
8	AUFO8	21.47	4.03	Fusiform	6.48	2.36	Oval	7.09	Round
9	AUFO9	18.66	4.08	Fusiform	5.71	2.07	Cylindrical	7.31	Round
10	AUFO10	14.17	3.35	Fusiform	7.03	2.96	Oval	8.56	Round
11	AUFO11	16.03	3.74	Cylindrical	6.68	2.37	Cylindrical	8.71	Oval
12	AUFO12	17.56	3.81	Cylindrical	7.36	1.06	Oval	9.03	Round
13	AUFO13	19.97	2.21	Cylindrical	5.31	1.97	Oval	9.87	Oval
14	AUFO14	21.03	2.71	Cylindrical	5.19	1.68	Cylindrical	9.26	Round
15	AUFO15	20.94	4.36	Fusiform	4.07	1.40	Oval	7.95	Oval
16	AUFO16	17.89	3.50	Fusiform	4.93	2.14	Oval	8.74	Oval
17	AUFO17	23.47	3.23	Fusiform	7.87	1.71	Cylindrical	8.30	Round
18	AUFO18	15.45	2.69	Fusiform	6.02	2.29	Oval	7.62	Round
19	AUFO19	16.21	2.56	Fusiform	4.73	1.11	Oval	7.23	Round
20	AUFO20	17.86	3.33	Fusiform	4.03	1.74	Oval	7.07	Round



Fig. 2. Microscopic view of A) Macroconidia (red arrow) and microconidia (blue arrow) and B) chlamydospores

seven-day-old Microscopic examination of cultures (Table 4 and Fig. 2) revealed variation in size macroconidia, and shape of chlamydospores. microconidia, and Macroconidia lengths ranged from 13.28 to 23.47 um. and breadths from 2.21 to 4.47 um. predominantly fusiform to cylindrical in shape. Microconidia measured between 4.03 and 7.87 um in length and 1.02 to 2.96 µm in breadth, displaying mainly oval to cylindrical morphologies. Chlamydospore size ranged from 7.07 to 9.87 µm, with shapes varying between oval and round across the isolates. The size range and shapes of macroconidia (fusiform to cylindrical), microconidia (oval to cylindrical), and chlamydospores (round to oval) were within the limits reported for *F. oxysporum* species.

These microscopic features serve as important taxonomic markers for distinguishing *F. oxysporum* from other fusaria and for identifying intraspecific variation (Garzón-Nivia et al., 2025; Leslie & Summerell, 2006). Moreover, the

presence and morphology of chlamydospores can be linked to the survival and persistence of the pathogen in soil, especially under adverse conditions.

3.3 Pathogenicity of *Fusarium oxysporum* isolates under pot culture condition

A pathogenicity assay was conducted to evaluate the virulence of 20 Fusarium oxysporum isolates (AUFO1-AUFO20) based on their ability to induce internal and external symptoms of vascular wilt in banana (Table 5). Among the tested isolates, AUFO5 exhibited the highest pathogenic potential, recording a mean internal score of 4.3 (PVWI- 87%) and a mean external score of 4.7 (PWI- 93%). This was followed by AUFO4 with internal and external scores of 4.0 (PVWI- 80%) and 4.3 (PWI- 87%), respectively. The least virulent was recorded in AUFO17 with both internal and external scores 0.3 and corresponding PVWI and PWI values of 7%, suggesting very low disease-causing ability.

Table 5. Pathogenicity of Fusarium oxysporum isolate under pot culture condition

S.No	Isolate	Mean internal score	PVWI	Mean external score	PWI
1	AUFO1	3.3	67	3.7	73
2	AUFO2	3.7	73	4.0	80
3	AUFO3	2.3	47	2.7	53
4	AUFO4	4.0	80	4.3	87
5	AUFO5	4.3	87	4.7	93
6	AUFO6	1.3	27	1.3	27
7	AUFO7	1.0	20	1.0	20
8	AUFO8	3.0	60	3.0	60
9	AUFO9	2.7	53	2.7	53
10	AUFO10	2.7	53	3.0	60
11	AUFO11	3.0	60	3.3	67
12	AUFO12	2.0	40	2.3	47
13	AUFO13	2.7	53	3.0	60
14	AUFO14	2.3	47	2.7	53
15	AUFO15	2.0	40	2.0	40
16	AUFO16	1.3	27	1.7	33
17	AUFO17	0.3	7	0.3	7
18	AUFO18	0.7	13	0.7	13
19	AUFO19	2.0	40	2.0	40
20	AUFO20	1.7	33	2.0	40

The existence of both highly aggressive and weakly pathogenic isolates within the same supports the hypothesis population pathogenicity in F. oxysporum is polyphyletic and influenced by the acquisition of mobile pathogenicity chromosomes and effector genes. The differential response observed among the isolates may also be linked to their ability to colonize host vascular tissues, produce toxins, or evade host defence mechanisms. These factors, together with host genotype and environmental conditions, contribute to disease expression under both field and pot culture conditions (Jenner & Henry, 2022; Nirmaladevi et al., 2016).

3.4 Molecular characterization of *F. oxysporum*

In the present study, PCR amplification using the universal primer pair ITS4/ITS5 vielded amplicons of approximately 500 bp, confirming the identity of the isolates as Fusarium oxysporum. Sequence analysis of all 20 isolates further validated their identity as F. oxysporum, oxysporum f. cubense, and sp. odoratissimum through **BLAST** searches **NCBI** conducted on the platform (www.blast.ncbi.nlm.nih.gov/Blast). The obtained sequences were submitted to GenBank, and accession numbers were assigned (Table 6 and Fig. 3). BLAST analysis revealed a high degree of similarity with sequences belonging to the F.

oxysporum clade. A phylogenetic tree illustrating the evolutionary relationships among the isolates was constructed using the neighbourioining method. Amplification of the translation elongation factor 1-alpha (TEF-1α) using specific primers yielded a clear and distinct product, which was subsequently submitted to the NCBI GenBank under the accession number PX241531. Race-specific PCR analysis revealed amplification only with the FOC R1 primer set, while no amplification was observed with the FOC R4 primer set, thereby confirming the isolate as belonging to Race 1 lineage of Fusarium oxysporum f. sp. cubense.

The high sequence identity with known entries in the *F. oxysporum* clade reinforces the taxonomic assignment and underlines the efficacy of ITSbased molecular tools in fungal diagnostics. The construction of a phylogenetic tree using the neighbour-joining method provided insights into evolutionary relationships among The clustering pattern observed isolates. corroborates the BLAST results and highlights the genetic diversity within the F. oxysporum complex. Phylogenetic grouping is especially relevant in the context of disease epidemiology and resistance breeding, as different lineages may vary in virulence, host range, and geographic distribution (Bibi et al., 2024; Hiremani & Dubey, 2019).

Table 6. Molecular profiling of Fusarium isolates

S. No	Name	Accession number	Scientific name
1	AUFO1	PV336003	Fusarium oxysporum f. sp. cubense
2	AUFO2	PV336085	Fusarium oxysporum
3	AUFO3	PV336130	Fusarium oxysporum
4	AUFO4	PV339469	Fusarium oxysporum
5	AUFO5	PV691490	Fusarium oxysporum f. sp. cubense
6	AUFO6	PV686885	Fusarium oxysporum
7	AUFO7	PV686889	Fusarium oxysporum
8	AUFO8	PV686892	Fusarium oxysporum f. sp. cubense
9	AUFO9	PV687400	Fusarium oxysporum
10	AUFO10	PV691491	Fusarium oxysporum f. sp. cubense
11	AUFO11	PV686883	Fusarium oxysporum f. sp. cubense
12	AUFO12	PV691492	Fusarium oxysporum
13	AUFO13	PV687424	Fusarium odoratissimum
14	AUFO14	PV687425	Fusarium oxysporum
15	AUFO15	PV687427	Fusarium oxysporum
16	AUFO16	PV687437	Fusarium oxysporum f. sp. cubense
17	AUFO17	PV687569	Fusarium oxysporum f.sp. cubense
18	AUFO18	PV738583	Fusarium oxysporum f. sp. cubense
19	AUFO19	PV738585	Fusarium oxysporum f. sp. cubense
20	AUFO20	PV738644	Fusarium oxysporum

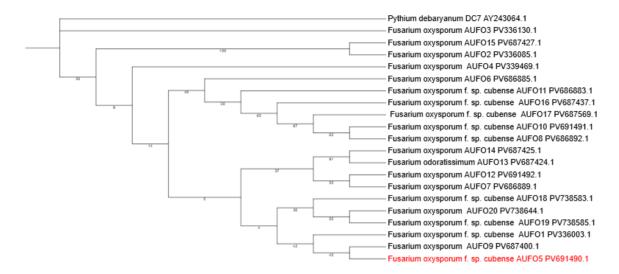


Fig. 3. Phylogeny tree based on ITS sequencing

4. CONCLUSION

The present study demonstrated considerable variation in Fusarium wilt incidence among banana cultivars and across different regions of Tamil Nadu. Morphological, pathogenicity, and molecular investigations confirmed occurrence of genetically diverse populations of Fusarium oxysporum f. sp. cubense (Foc) exhibiting variable virulence. These findings underscore the urgent need for deploying resistant cultivars and implementing regionmanagement practices to specific reduce pressure. In addition, the disease study emphasizes the importance of continuous molecular surveillance for early detection of emerging virulent lineages and monitoring pathogen evolution. Integration of resistant varieties with good agronomic practices, soil health management, and biological control strategies could form the cornerstone of sustainable Fusarium wilt management. Moreover, development of rapid molecular diagnostic tools and establishment of a regional pathogen monitoring network would further to prevent strengthen efforts large-scale epidemics and ensure the long-term sustainability of banana cultivation in Tamil Nadu.

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- 1. ChatGPT was used to rephrase sentences into academic style.
- 2. ChatGPT was used to improve grammatical accuracy.
- ChatGPT was used to refine fluency and readability of the manuscript.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

Abdoussalami, A., Hu, Z., Islam, A. R. M. T., & Wu, Z. (2023). Climate change and its impacts on banana production: A systematic analysis. *Environment, Development and Sustainability, 25,* 12217–1246. https://doi.org/10.1007/s10668-023-03168-2

Bibi, A., Mubeen, F., Rizwan, A., Ullah, I., Hammad, M., Waqas, M. A. B., et al. (2024). Morpho-molecular identification of *Fusarium equiseti* and *Fusarium oxysporum* associated with symptomatic wilting of potato from Pakistan. *Journal of Fungi*, 10, 701. https://doi.org/10.3390/jof10100701

Cha, S.-D., Jeon, Y.-J., Ahn, G.-R., Han, J.-I., Han, K.-H., & Kim, S.-H. (2007).

- Characterization of *Fusarium oxysporum* isolated from paprika in Korea. *Mycobiology*, 35, 91–96. https://doi.org/10.4489/MYCO.2007.35.2.0
- Chukwu, S. C., Awala, S. K., Angombe, S., Valombola, J. S., Nanhapo, P. I., Mberama, C., et al. (2025). Recent progress in tissue culture techniques and biotechnological innovations for banana production (*Musa spp.*): A review. *Discover Plants*, 2, 13. https://doi.org/10.1007/s44372-025-00099-2
- Costa, T. F., Santos, M. C., De Souza Junior, L. C., Brito, D. A., De Jesus Rocha, A., Lino, L. S. M., et al. (2025). Gamma radiation-induced mutagenesis in the development of Cavendish subgroup banana cultivars resistant to *Fusarium oxysporum* f. sp. *cubense*. *Euphytica*, 221, 147. https://doi.org/10.1007/s10681-025-03595-4
- Dhanyasree, K., Rafeekher, M., & Premachandran, A. (2022). Abiotic stress management in fruit crops: A review. *AG*. https://doi.org/10.18805/ag.R-2557
- Dita, M., Barquero, M., Heck, D., Mizubuti, E. S. G., & Staver, C. P. (2018). Fusarium wilt of banana: Current knowledge on epidemiology and research needs toward sustainable disease management. Frontiers in Plant Science, 9, 1468. https://doi.org/10.3389/fpls.2018.01468
- Esquivel Chávez, F., Campos-Avelar, I., Chávez Luzanía, R. A., Montoya-Martínez, A. C., Parra-Cota, F. I., & De Los Santos Villalobos, S. (2025). Biotic stresses of horticultural crops: Present and future challenges. In *Biotic Stress Tolerance in Horticultural Crops* (pp. 19–32). Elsevier. https://doi.org/10.1016/B978-0-443-27324-7.00002-1
- Garzón-Nivia, M. A., Mártiz Mártiz, J., Moya-Elizondo, E. A., Ruiz, B., Cornejo, J. C., & Valdés-Gómez, H. A. (2025). Characterization and identification of Neocosmospora solani and Fusarium oxysporum causing root necrosis and wilting of orange trees in Chile. Plants, 14, 376.
 - https://doi.org/10.3390/plants14030376
- Harish, J., Jambhulkar, P. P., Bajpai, R., Arya, M., Babele, P. K., Chaturvedi, S. K., et al. (2023). Morphological characterization, pathogenicity screening, and molecular identification of *Fusarium* spp. isolates

- causing post-flowering stalk rot in maize. *Frontiers in Microbiology*, *14*, 1121781. https://doi.org/10.3389/fmicb.2023.112178
- Hiremani, & N. S., Dubey, S. (2019). Phylogenetic relationship among Indian population of Fusarium oxysporum f. sp. lentis infectina lentil and development of specific SCAR markers for detection. 3 Biotech, 9, https://doi.org/10.1007/s13205-019-1734-4
- Ismaila, A. A., Ahmad, K., Siddique, Y., Wahab, M. A. A., Kutawa, A. B., Abdullahi, A., et al. (2023). *Fusarium* wilt of banana: Current update and sustainable disease control using classical and essential oils approaches. *Horticultural Plant Journal*, 9, 1–28.
 - https://doi.org/10.1016/j.hpj.2022.02.004
- Jenner, B. N., & Henry, P. M. (2022). Pathotypes of Fusarium oxysporum f. sp. fragariae express discrete repertoires of accessory genes and induce distinct host transcriptional responses during root infection. Environmental Microbiology, 24, 4570–4586. https://doi.org/10.1111/1462-2920.16101
- Kumari, P., Gaur, S. S., & Tiwari, R. K. (2023).

 Banana and its by-products: A comprehensive review on its nutritional composition and pharmacological benefits. *eFood*, *4*, e110. https://doi.org/10.1002/efd2.110
- Leslie, J. F., & Summerell, B. A. (2006). *The Fusarium laboratory manual*. Ames (lowa): Blackwell Publishing.
- Maseko, K. H., Regnier, T., Meiring, B., Wokadala, O. C., & Anyasi, T. A. (2024). *Musa* species variation, production, and the application of its processed flour: A review. *Scientia Horticulturae*, 325, 112688.
 - https://doi.org/10.1016/j.scienta.2023.1126
- Nirmaladevi, D., Venkataramana, M., Srivastava, R. K., Uppalapati, S. R., Gupta, V. K., Yli-Mattila, T., et al. (2016). Molecular phylogeny, pathogenicity and toxigenicity of Fusarium oxysporum f. sp. lycopersici. Scientific Reports, 6, 21367. https://doi.org/10.1038/srep21367
- Olivares, B. O., Rey, J. C., Lobo, D., Navas-Cortés, J. A., Gómez, J. A., & Landa, B. B. (2021). *Fusarium* wilt of bananas: A review of agro-environmental factors in the Venezuelan production system affecting its

- development. *Agronomy*, 11, 986. https://doi.org/10.3390/agronomy11050986
- Pareek, S. (2016). Nutritional and biochemical composition of banana (*Musa spp.*) cultivars. In *Nutritional Composition of Fruit Cultivars* (pp. 49–81). Elsevier. https://doi.org/10.1016/B978-0-12-408117-8.00003-9
- Pérez-Vicente, L., & Dita, M. A. (2014). Fusarium wilt of banana or Panama disease by Fusarium oxysporum f. sp. cubense: A review on history, symptoms, biology, epidemiology and management. In Pérez-Vicente, L., Dita, M. A., & Martínez-de la Parte, E. (Eds.), Technical Manual Prevention and Diagnostic of Fusarium Wilt (Panama Disease) of Banana Caused by Fusarium oxysporum f. sp. cubense Tropical Race 4 (TR4) (pp. 5–30). FAO. https://www.scirp.org/reference/references papers?referenceid=1454141
- Ranjha, M. M. A. N., Irfan, S., Nadeem, M., & Mahmood, S. (2022). A comprehensive review on nutritional value, medicinal uses, and processing of banana. *Food Reviews International*, 38, 199–225. https://doi.org/10.1080/87559129.2020.172 5890
- Sasaki, N., Yokoi, S., Hidalgo, C. X. T., Suzuki, S., Takahashi, S., Sha, K., et al. (2025). Impact and control of transboundary and invasive banana (*Musa spp.*) wilt pathogen, *Fusarium oxysporum* f. sp. *cubense*. In Nagamine, T., Masuda, M., & Irie, K. (Eds.), *Plant Genetic Resources (PGR) for Sustainable Crop Production* (pp. 121–130). Springer Nature Singapore. https://doi.org/10.1007/978-981-96-7117-5 11
- Thangavelu, R., Amaresh, H., Gopi, M., Loganathan, M., Nithya, B., Ganga Devi,

- P., et al. (2024). Geographical distribution, host range and genetic diversity of *Fusarium oxysporum* f. sp. *cubense* causing *Fusarium* wilt of banana in India. *Journal of Fungi*, 10, 887. https://doi.org/10.3390/jof10120887
- Thangavelu, R., Loganathan, M., Arthee, R., Prabakaran, M., & Uma, S. (2020). Fusarium wilt: A threat to banana cultivation and its management. CABI Reviews, 1–24. https://doi.org/10.1079/PAVSNNR2020150 04
- Thangavelu, R., Mostert, D., Gopi, M., Devi, P. G., Padmanaban, B., Molina, A. B., et al. (2019). First detection of *Fusarium oxysporum* f. sp. *cubense* tropical race 4 (TR4) on Cavendish banana in India. *European Journal of Plant Pathology*, 154, 777–786. https://doi.org/10.1007/s10658-019-01701-6
- Tinzaara, W., Mutambuka, M., Oyesigye, E., Blomme, G., Dita, M., Gold, C. S., et al. (2024). Banana wilt diseases: Current status and future research strategies for their management. *International Journal of Pest Management*, 70, 290–309.
 - https://doi.org/10.1080/09670874.2021.199 2685
- Vieira, B. M., De Mello E Silva, G. N., & Silva, M. I. (2025). Nutritional elements II: Vitamins and minerals. In Fajemiroye, J. O. (Ed.), Fundamentals of Drug and Non-Drug Interactions (pp. 57–86). Springer Nature Switzerland. https://doi.org/10.1007/978-3-031-80107-5_3
- Vignesh, M., Selvakumar, R., & Azhagesan, R. (2023). Marketing strategy and performance of banana in Kanniyakumari district of Tamil Nadu.

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