

# PHYSIOLOGICAL STUDIES OF *FUSARIUM OXYSPORUM* F. SP. *CUBENSE* CAUSING PANAMA WILT IN BANANA

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## KEYWORDS

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## ABSTRACT

Panama wilts of banana caused *Fusarium oxysporum* f.sp. *cubense* is an important and destructive disease of Karnataka and many other banana growing states of India. The effect of temperature, pH levels and light spectra on mycelial growth of *Fusarium oxysporum* f.sp. *cubense* was investigated. The fungus grew at temperatures ranging from 15 to 35°C, with optimum growth and sporulation at 25°C and least growth and sporulation was observed at 15 and 35°C. The most suitable pH level for the growth was ranged from 5-7 with respect different isolates. The growth was reduced under different light spectra when compared to alternate cycles of light and darkness.

## INTRODUCTION

Karnataka is known for high quality bananas especially 'Nanjangudrasabale' (NRB) is an elite native variety of banana originated from a place called 'Nanjangud'. It is popular and highly priced fruit in Karnataka and in other states because of its special qualities such as taste, aroma, colour, fibrous texture and nutritional content. As we know, the panama wilt of banana is a major constrain of successful production of this crop. *Fusarium oxysporum* f.sp. *cubense* (Smith, 1910; Snyder and Hansen, 1940), is one of the most destructive soil borne vascular wilt pathogen especially race1, is an important disease on Rasthali group (AAB) of banana. In southern India, incidence of Panama wilt is widespread in some districts the disease incidence is as high as 80-90% (Sivamani, 1987).

Panama disease in banana is one of the most harmful diseases that have caused the greatest damage to this crop throughout the 20th century. The highly susceptible cultivar, Gross Michel, used by export trades until about 1960, was replaced by clones of the Cavendish Subgroup. Although Cavendish cultivars are resistant to Race 1 (FOC-1) and Race 2 (FOC-2), the Race 4 of the pathogen (FOC-4) damages these cultivars in subtropical banana-growing regions. The continued use of the Cavendish cultivars is now threatened in these regions and producers in the tropics, mindful of disastrous epidemics which occurred in Gross Michel, are concerned that Race 4 or a similar one might develop in their areas (Ploetz, 1990).

Panama wilt of banana is one of the internationally known disease but no much information is available on the

physiological requirements of *Fusarium oxysporum* f.sp. *cubense*. The present study was therefore undertaken to study the effect of temperature, pH and light spectra on mycelial growth of *Fusarium oxysporum* f. sp. *cubense* which will be helpful in management strategy.

## MATERIALS AND METHODS

An *in vitro* experiment was conducted during 2011 at K. R. C. College of Horticulture, Arabhavi to find out the suitable temperature, light and pH for the growth and sporulation of *Fusarium oxysporum* f. sp. *cubense*. The experiment was designed in Complete Randomized Design (CRD).

### Temperature

The different temperature tried for growth and sporulation of the pathogen were 15, 20, 22, 25, 28, 30 and 35°C. Thirty ml of sterilized PDA media was dispensed in 90 mm diameter petridishes and incubated aseptically with 5 mm disc of the pathogen from a seven days old culture. Petridishes were incubated at different temperature and each treatment was replicated three times. At the end of seventh and ninth day of incubation, observation on colony diameter was recorded and after ninth day of incubation sporulation was recorded. (Khilare and Ahmed, 2012).

### Light

Effect of light on growth and sporulation of the pathogen was studied on PDA media by exposing the pure culture to continuous light, continuous dark, alternating with 12 hours

complete light and 12 hours complete darkness along with control (under normal room condition). The inoculation of culture to petridishes containing PDA media was done as explained earlier with five replication per treatment. The plates were incubated at  $26 \pm 1^\circ\text{C}$  for nine days. Observation on colony diameter and sporulation were recorded. Sharma et al. (2005)

#### Hydrogen ion concentration (pH)

Potato dextrose broth was used as a basal medium. pH of the liquid medium was adjusted by using 0.1N alkali (NaOH) or 0.1N acid (HCl). The pH of the medium used were 4.0, 5.0, 6.0, 7.0, 8.0 and 9.0. The culture was inoculated to each of 100 ml flask containing 30 mL of basal medium and incubated at  $26 \pm 1^\circ\text{C}$  for ten days. Four replications were maintained in each treatment. Dry mycelial weight of the fungus was recorded. Results were analyzed statistically. (Khilare and Ahmed, 2012).

## RESULTS AND DISCUSSION

The result of the present study on temperature, light and hydrogen ion concentration as well as relevant discussions has been presented under following sub heads.

#### Effect of temperature on the growth and sporulation of *Fusarium oxysporum* f. sp. *cubeuse*

Pathogen was inoculated on potato dextrose agar medium as described in material and methods and incubated at different temperatures of 15, 20, 22, 25, 28, 30 and  $35^\circ\text{C}$  for nine

**Table 1: Effect of temperature on the growth and sporulation of *Fusarium oxysporum* f. sp. *cubeuse***

Temperature ( $^\circ\text{C}$ )	Colony diameter (mm)		Sporulation at 9 <sup>th</sup> day
	7 <sup>th</sup> day	9 <sup>th</sup> day	
15	49.67	60.33	+++
20	56.33	68.67	+++
22	60.67	73.33	+++
25	74.33	86.33	++++
28	71	82	++++
30	68.67	77	++++
35	22.67	27.33	++
S.Em $\pm$	1.65	1.69	
CD @ 1%	6.94	7.1	

++++ = >75 conidia per microscopic field; +++ = 50-75 conidia per microscopic field; ++ = 25-50 conidia per microscopic field; + = 1-25 conidia per microscopic field.

**Table 2: Effect of light and darkness on the growth and sporulation of *Fusarium oxysporum* f. sp. *cubeuse***

Treatment	Colony diameter (mm)		Sporulation at 9 <sup>th</sup> day
	7 <sup>th</sup> day	9 <sup>th</sup> day	
Continuous light	61	70.8	+++
Continuous dark	60	65.2	+++
12 hours light and 12 hours dark	66	77.4	++++
Control	59.2	71.2	++++
S.Em $\pm$	1.09	1.41	
CD @ 1%	4.5	5.83	

++++ = >75 conidia per microscopic field; +++ = 50-75 conidia per microscopic field; ++ = 25-50 conidia per microscopic field; + = 1-25 conidia per microscopic field

days. The average radial growth and sporulation were recorded and are presented in Table 1 and Figure 1.

The results of the study indicated that, the maximum growth of the fungus was 86.33 mm at  $25^\circ\text{C}$  followed by 82.00 mm at  $28^\circ\text{C}$ , 77.00 mm at  $30^\circ\text{C}$ , 73.33 mm at  $22^\circ\text{C}$ , 68.67 mm at  $20^\circ\text{C}$  and 60.33 mm at  $15^\circ\text{C}$ . The least growth of fungus was 27.33 mm at  $35^\circ\text{C}$ , which differed significantly from the growth at other temperatures.

Heavy sporulation was recorded when the plates were incubated at 25, 28 and  $30^\circ\text{C}$  temperature. However, sporulation was moderate at 15, 20 and  $22^\circ\text{C}$  and it was least at  $35^\circ\text{C}$ .

Studies on the influence of temperature on growth of *Fusarium oxysporum* f. sp. *cubeuse* showed maximum growth at  $25^\circ\text{C}$  followed by  $28^\circ\text{C}$ . Whereas, optimum temperature range was  $20-30^\circ\text{C}$ . Similar experiment was conducted by Farooq et al. (2005) who observed that temperature of  $25^\circ\text{C}$  and  $30^\circ\text{C}$  were the best for *Fusarium oxysporum* f. sp. *ciceri* were it has attained maximum growth. Khan et al. (2011) reported that temperature level of  $30^\circ\text{C}$  was the common optimum for growth of *Fusarium oxysporum* f. sp. *ciceri* followed by  $25^\circ\text{C}$ . Ramteke and Kamble (2011) reported that growth of *Fusarium solani* was good at  $20^\circ\text{C}$  and  $30^\circ\text{C}$  and it was relatively low at  $10^\circ\text{C}$  and  $35^\circ\text{C}$ . Khilare and Ahmed (2012) reported that temperature from 25 to  $35^\circ\text{C}$  were most favorable for the growth of *Fusarium oxysporum* f. sp. *cicer*. Gupta et al. (2010) reported optimum temperature for growth of *Fusarium* spp. isolates was  $28^\circ\text{C}$ . Bhale (2012) results revealed that radial growth and spore germination of *F. oxysporum* f. sp. *spinaciae* was maximum temperature at  $25^\circ\text{C}$ .

#### Effect of light on the growth and sporulation of *Fusarium oxysporum* f. sp. *cubeuse*

An experiment was conducted to study the effect of light and darkness on the growth and sporulation of *Fusarium oxysporum* f. sp. *cubeuse*. The fungus was inoculated on sterilized potato dextrose agar medium as described in material and methods and the plates were exposed to alternate cycles of 12 hours light and 12 hours darkness, continuous light and continuous dark for 24 hours along with control (under normal room condition) for nine days. The results are presented in Table 2 and Figure 2.

The maximum growth of the fungus (77.40 mm) was recorded in 12 hours light and darkness followed by 70.80 mm in continuous light. Least growth of the fungus (65.20 mm) was recorded when exposed to continuous dark.

Heavy sporulation was recorded when culture was exposed to alternate cycle of 12 hours light and 12 hours darkness. Good sporulation was recorded when culture was exposed under normal room condition. Moderate sporulation was noticed when culture was exposed to continuous light and dark conditions.

The studies carried out in the present investigation with *Fusarium oxysporum* f. sp. *cubeuse* exhibited the maximum growth and sporulation, when inoculated plates were exposed to alternate light and dark conditions (12 hours light alternated with 12 hours dark) followed by continuous light and continuous dark. Similar observations were recorded by Sharma et al. (2005) who found that the growth and

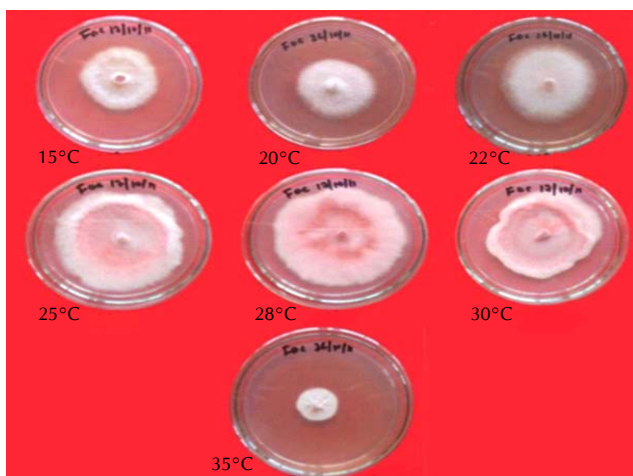


Figure 1: Effect of temperature on growth of *Fusarium oxysporum* f. sp. *cubense*

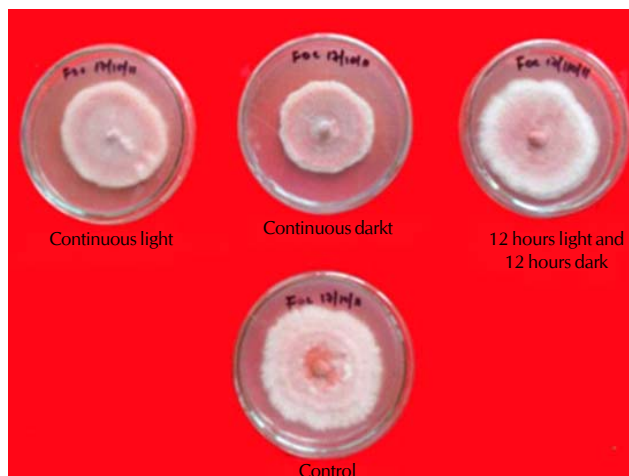
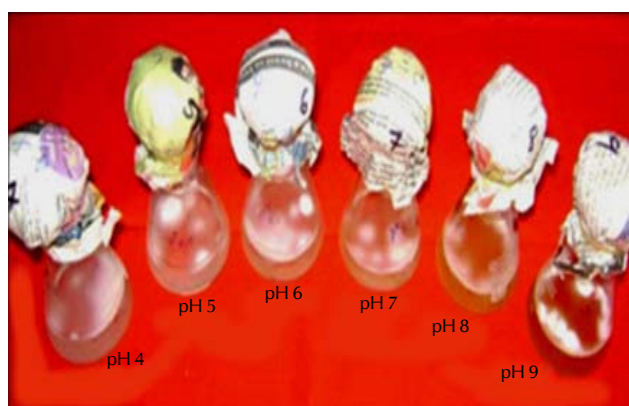


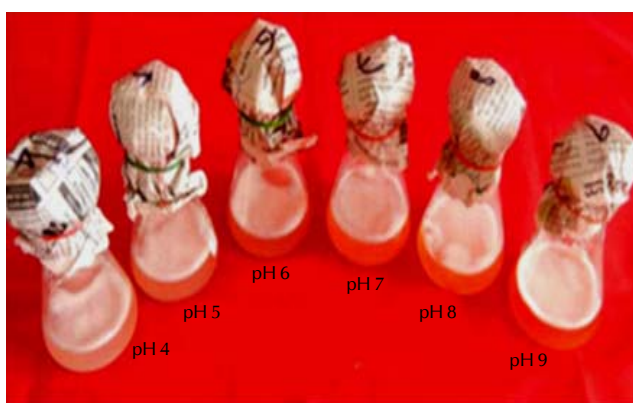
Figure 2: Effect of light on growth of *Fusarium oxysporum* f. sp. *cubense*



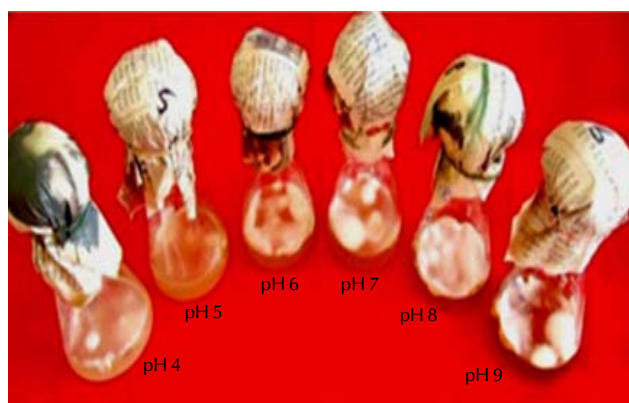
Bangalore isolate



Channapatna isolate



Mandya isolate



Mysore isolate

Figure 3: Effect of different pH level on growth of different isolates of *Fusarium oxysporum* f. sp. *cubense* on potato dextrose broth

sporulation of *Fusarium oxysporum* f. sp. *lini* was excellent at alternate cycles of 12 hours each of light and darkness. Bhale (2012) reported that continuous light and white light were found ideal for maximum radial growth and spore germination of *F. oxysporum* f. sp. *spinaciae*.

**Effect of hydrogen ion concentration (pH) on the growth and sporulation of *Fusarium oxysporum* f. sp. *cubense***

Four isolates of *Fusarium oxysporum* f. sp. *cubense* were incubated at  $26 \pm 1^\circ\text{C}$  for nine days maintained at different pH of 4.0, 5.0, 6.0, 7.0, 8.0 and 9.0. The mean dry weight of mycelium was recorded and present in Table 3 and Figure 3. The variation in growth among the isolates at different pH were found to be significant. Result of the study revealed that Bangalore isolate produced maximum growth of 628.48 mg

**Table 3: Effect of pH on the growth of different isolates of *Fusarium oxysporum* f. sp. *cubense* on potato dextrose broth**

pH	Mean dry weight of mycelium (mg)			
	Bangalore isolate	Channapatna isolate	Mandya isolate	Mysore isolate
4.0	483.38	309.00	702.10	365.33
5.0	586.13	232.70	835.88	468.63
6.0	628.48	421.05	776.00	634.63
7.0	514.20	446.73	728.70	629.18
8.0	473.08	364.48	717.23	471.53
9.0	448.03	321.78	659.60	430.13
S.Em±	1.66	1.30	2.56	1.11
CD @ 1%	6.74	5.29	10.41	4.50

at pH 6.0 followed by 586.13 mg at pH 5.0, 514.20 mg at pH 7.0, 483.30 mg at pH 4.0 and 473.08 mg at pH 8.0. Least growth of 448.03 mg was observed at pH 9.0.

Channapatna isolate produce maximum mycelial growth at pH 7.0 (446.73 mg) followed by 421.05 mg at pH 6.0, 364.48 mg at pH 8.0, 321.78 mg at pH 9.0, 309.00 mg at pH 4.0 and growth was declined at pH 5.0 (232.70 mg).

Mandya isolate produced maximum mycelial growth of 835.88 mg at pH 5.0 followed by 776.00 mg in pH 6.0, 728.70 mg at pH 7.0, 717.23 mg at pH 8.0, 702.10 mg at pH 4.0 and growth was reduced at pH 9.0 (659.60 mg).

At pH 6.0 Mysore isolate produced maximum mycelial growth of 634.63 mg followed by 629.18 mg at pH 7.0, 471.53 mg at pH 8.0, 468.63 mg at pH 5.0 and 430.13 mg at pH 9.0. Lowest growth of 365.33 mg was found at pH 4.0.

The isolates of *Fusarium oxysporum* f. sp. *cubense* could grow under a wide range of hydrogen ion concentrations from 4.0 to 9.0. The growth of all the four isolates was minimum at pH 4.0 increased till pH 7.0. The optimum pH range was 5.0 to 7.0 in all the four isolates. The present findings are in agreement with the reports of Farooq *et al.* (2005) reported that maximum growth of *Fusarium oxysporum* f. sp. *ciceri* was at pH 7. Naik *et al.* (2010) reported that the most suitable pH level for growth of *Fusarium oxysporum* f. sp. *vanillae* was 5.0 and 6.0. Isolates of Fov-3 and Fov-6 showed highest growth of 62.4 and 62.1 mm respectively. The maximum growth of *Fusarium oxysporum* f. sp. *psidii* was recorded when the pH was at the level of 5.5 (1208 mg) followed by a pH of 5.0 (956 mg) and then pH 6.0 (953 mg) (Gupta *et al.*, 2010). Khan *et al.* (2011) found that maximum growth of mycelial mat of *Fusarium oxysporum* f. sp. *ciceri* was recorded at pH 7.0 and was significantly superior to other pH levels followed by pH 6.5 and at pH 6.0. Khilare and Ahmed (2012) reported that effect of pH was highest at six with sporulation of 24.70 conidia/ml.

However, it was found that the range from 4.5 to 8.0 is suitable for the growth of *Fusarium oxysporum* f. sp. *ciceri*. Bhale (2012) reported that maximum radial growth and spore germination of *F. oxysporum* f. sp. *spinaciae* was found at 6.5, followed by 7.5 pH. Jaruhar and Prasad (2011) reported that pH level 6.0 was found optimum for the growth as well as sporulation of the *Fusarium oxysporum* schlecht. f. sp. *lentis*. Sporulation of chlamydo-spore was however found best in the pH level 4.0.

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