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Physiological studies of *Colletotrichum musae* the causal agent of Anthracnose disease of banana

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ABSTRACT

Effect of different temperature, light intensity and pH were tested against the growth and sporulation of *Colletotrichum musae* under *in vitro* conditions. Results indicated that the growth of *Colletotrichum musae* was maximum at 30 ^{0}C (72.25 mm) followed by 25 ^{0}C (68.25 mm), 20 ^{0}C (53.00 mm), 15 ^{0}C (52.75 mm) and it was lowest growth (12.00 mm) at 35 ^{0}C. Exposure of *Colletotrichum musae* to alternate cycles of 12 hr light and 12 hr darkness, continous light and under normal condition (room temperature) resulted in the maximum mycelial growth (90.00 mm) and heavy sporulation. The variation in growth of *Colletotrichum musae* at different pH were found to be significant. Result of the study revealed that at pH 7.0 fungus produced maximum growth of 977.0 mg followed by 960.0 mg at pH 8.0, 957.0 mg at pH 6.0, 948.0 mg at pH 5.0 and 922.0 mg at pH 4.0.

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INTRODUCTION

Banana (*Musa* sp.) is one of the important fruit crops of the world as well as India. It is a good source of energy, minerals and vitamins and is one of the biggest single trade items in international fruit trade (Snehalatharani and Khan, 2009). Banana is susceptible to several diseases resulting in massive and extensive postharvest losses during transportation and storage (Basel *et al*., 2002). Anthracnose caused by *Colletotrichum musae* (Berk. and Curt.) Arx. is one of the most important and widely distributed diseases of ripening and ripe bananas, and is particularly associated with wastage following injuries in the form of scratches and other wounds sustained by the fruits during handling and transport (Wardlaw, 1934). There are also losses in local markets because of disease infection of ripe fruits. The fungus can infect banana fruits at any time during the growing season in the field (Simmonds and Mitchell, 1940). Banana anthracnose usually starts as quiescent infections on green fruit in the field. However, successful penetration of the fungus is restricted by accumulation of phytoalexins as the fruits ripen (Jegger *et al*., 1995 and Turner, 1995). Therefore, symptoms generally can

be seen only in overripe fruits. Anthracnose becomes a serious problem when bananas are shipped as bunches for a long time and ripened under high temperature (Meredith, 1960a). *Colletotrichum musae* is the most important pathogen on wounded green and ripe banana fruits (Meredith, 1960b and Stover and Simmonds, 1987b). Occasionally, the fungus invades necks of green fingers when damaged by flexing (Wardlaw, 1995). Lesions are sunken and covered with salmon-colored acervuli (Sutton and Waterston, 1970 and Ranasinghe *et al*.,2005). Infections stimulate ripening of fruits and lesions elongate with ripening. On ripening fruits, sunken brown spots develop with orange acervuli (Stover and Simmonds, 1987). So, the present study is, therefore, conducted to examine the effect of temperature, light and pH for growth and sporulation of *Colleotrichum musae*.

MATERIAL AND METHODS

An experiment was carried out during 2013 at K. R. C. College of Horticulture, Arabhavi to evaluate the effect of temperature, light and pH for growth and sporulation of *Colletotrichum musae.*

Effect of temperature on the growth and sporulation of *Colletotrichum musae*:

The different temperatures tried for growth and sporulation of the pathogen were 15, 20, 25, 30 and 35°C. Twenty ml of sterilized PDA media was poured into 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 temperatures and each treatment was replicated four times. Observations on colony diameter and sporulation were recorded.

Effect of light on the growth and sporulation of *Colletotrichum musae:*

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 conditions).Twenty 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 light conditions and each treatment was replicated five times following Completely Randomized Design. Observations on colony diameter and sporulation were recorded.

Effect of hydrogen ion concentration (pH) on the growth of *Colletotrichum musae:*

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 28°C for ten days. Four replications were maintained in each treatment. Dry mycelial weight of the fungus was recorded. Results were analyzed statistically.

RESULTS AND DISCUSSION

Temperature plays an important role among the external factors which influence the growth and reproduction of fungi. Results revealed that, the maximum growth of the fungus was attained at 30 °C (72.25 mm) followed by 25 °C (68.25 mm), 20 °C (53.00 mm), 15 °C (52.75 mm) and it was lowest growth (12.00 mm) at 35 °C. At seven days the maximum growth of fungus was 90.00 mm at 30 °C followed by 89.25 mm at 25 °C, 72.25 mm at 20 °C, 66.75 mm at 15 °C. The least growth of fungus was 21.00 mm at 35 °C, which differed significantly from the growth at other temperatures. The maximum sporulation was recorded at 20 °C, 25 °C and 30 °C. Moderate and least sporulation were recorded at 15 °C and 35 °C, respectively. Similar experiment was conducted by Lim *et al.* (2002) who observed maximum mycelial growth of *Colletotrichum musae* ST-01 at 25-30 °C (Table 1). Vinod *et al.* (2009) reported that 30 °C is required for the good growth of *Colletotrichum gloeosporioides* causal agent of anthracnose of papaya. Thangamani *et al.* (2011) revealed that the highest mean radial mycelial growth of *Colletotrichum musae* was observed at 30°C which was followed by 25°C and 20°C.

Light has profound effect on growth and sporulation of fungi. At five days, the maximum growth of the fungus (90.00 mm) was recorded in 12 hours light and 12 hours dark, followed by continous light (89.00mm), under normal conditions (88.00 mm) and it was least when exposed to continous dark (70.20 mm).While at 7th day maximum growth of the fungus (90.00mm) was recorded in 12 hours light and darkness , continous light and under

normal conditions .The least growth of the fungus (78.80 mm) was recorded when exposed to continuous dark (Table 2).

Heavy sporulation was recorded when culture was exposed to alternate cycle of 12 hours light and 12 hours darkness, continous light and under normal condition. Good sporulation was recorded when culture was exposed under continous dark. Similar observations were recorded by Kamanna (1996); Ashoka (2005); Mesta (1996); Sudhakar (2000); Prashanth (2007) and Vinod *et al.* (2009). Venkataravanappa (2002), who reported the excellent growth and sporulation of *Colletotrichum gloeosporioides* at alternate cycles of 12 hours each of light and darkness. Thangamani *et al.* (2011) reported maximum mycelia growth and sporulation of *Colletotrichum musae* at alternate cycles of 12 hours each of light and darkness.

The variation in growth of *Colletotrichum musae*

Table 1 : Effect of temperature on the growth and sporulation of *Colletotrichum musae*

Temperature (^{0}C)	Colony diameter (mm)		
	5th day	7th day	Sporulation at 7th day
15 ^{0}C	52.75	66.75	+ + +
20 ^{0}C	53.00	72.25	+ + + +
25 ^{0}C	68.25	89.25	+ + + +
30 ^{0}C	72.25	90.00	+ + + +
35 ^{0}C	12.00	21.00	+ +
Control	73.50	89.62	+ + + +
S.E. ±	2.17	0.71	-
C.D. (P=0.01)	8.85	2.91	-
CV(%)	7.86	2.00	-

++++ = >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 *Colletotrichum musae*

Treatments	Colony diameter (mm)		
	7th day	9th day	Sporulation at 9th day
Continuous light	89.00	90.00	+ + + +
Continuous dark	70.20	78.80	+ + +
12 hours light and 12 hours dark	90.00	90.00	+ + + +
Control	88.00	90.00	+ + + +
S.E. ±	1.86	0.18	-
C.D. (P=0.01)	7.71	0.77	-
CV (%)	4.96	0.47	-

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

Table 3: Effect of pH on the growth of *Colletotrichum musae* in potato dextrose broth

pH	Mean dry weight of mycelium(mg)
4	922.0
5	948.0
6	957.0
7	977.0
8	960.0
9	907.0
S.E. ±	0.40
C.D. (P=0.01)	1.66

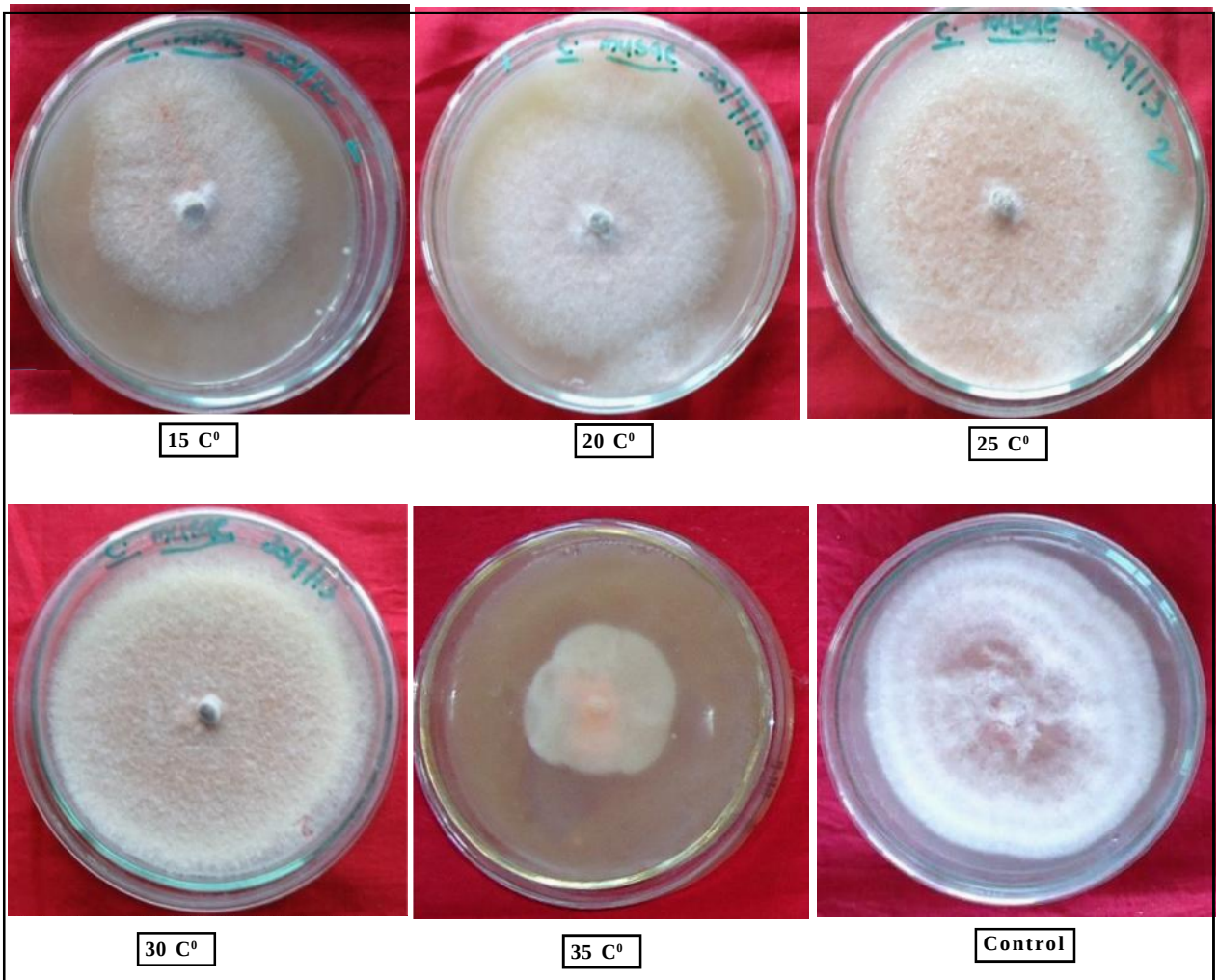

Plate 1: Effect of temperature on growth of *Colletorichum musae*

Plate 2 : Effect of light and darkness on the growth of *Colletorichum musae*

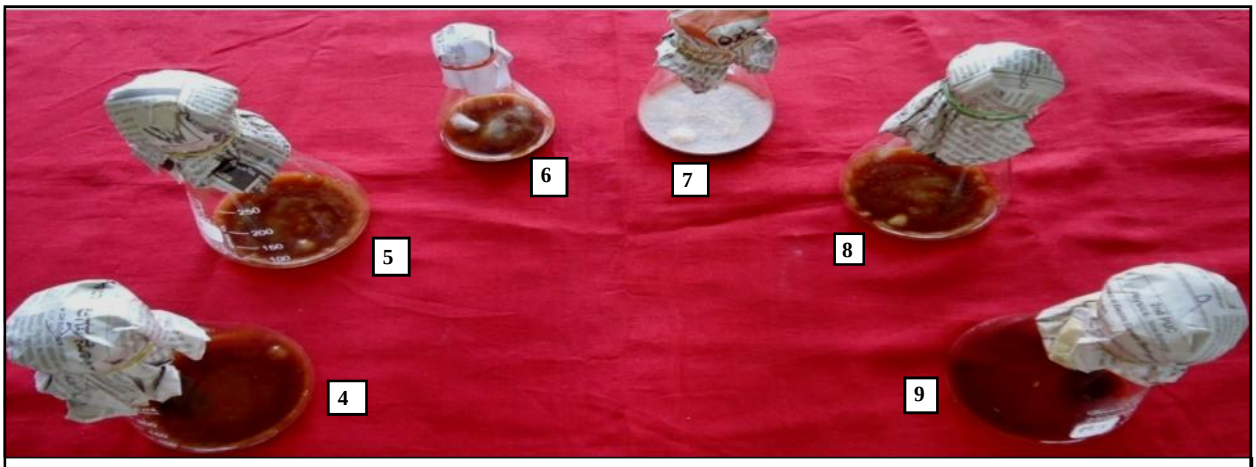

Plate 3 : Dry weight of Colletotrichum musae at different PH levels in potato dextrose broth

at different pH were found to be significant. Result of the study revealed that at pH 7.0 fungus produced maximum growth of 977.0 mg followed by 960.0 mg at pH 8.0, 957.0 mg at pH 6.0, 948.0 mg at pH 5.0, 922.0 mg at pH 4.0. Least growth of 907.0 mg was observed at pH 9.0. The present findings are in agreement with the reports of Lim *et al.* (2002) who reported pH 5.5-7.0 as the optimum pH for mycelial growth of *Colletotrichum musae* . At this pH level, mycelial growth did not show much difference suggesting an insignificant relationship. Thangamani *et al*. (2011) observed maximum mean mycelia growth of *Colletotrichum musae* (87.52 mm) at pH 7.0 followed by pH 6.5 (85.97 mm) and pH 6.00 (79.85 mm). The lowest mean mycelial growth was recorded at pH 4.0 (34.54 mm). The pH below six and above seven was detrimental to the growth of pathogen.

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