Cultural studies of *Pestalotiopsis mangiferae* causing grey blight of mango under *in vitro* condition

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**Abstract**

The present investigation entitled “Cultural Studies of *Pestalotiopsis mangiferae* causing Grey Blight of Mango under *In vitro* condition”, was under taken to evaluate for various cultural studies of the given test fungus. The diseased samples were collected from different localities of Uttarkhand, i.e Dehradun, Haridwar and Rishikesh during the 2019-2020. The pathogen was isolated from the diseased samples and the pathogen was maintained in potato dextrose agar. The potential species of *Pestalotiopsis mangiferae* was identified and isolated. In the culture study of the pathogen was grown on five different solid media which were selected for this analysis. Observations on the radial growth rate of *P. mangiferae* in four replications were reported at the end of the 6th day of inoculation. Irrespective of the media used Czapek dox agar (CDA) was found to be the most suitable culture medium for the growth of *Pestalotiopsis mangiferae* effectively. Where Corn meal agar (CMA) has been found to be a non-preferable culture medium for the growth of *Pestalotiopsis mangiferae*.

**Keywords:** Mango, Grey Blight, *Pestalotiopsis mangiferae* mycelium growth, solid media, inhibition, potato dextrose agar, nutrient agar, cornmeal agar, sabourauds dextrose agar and Czapek’s dox

**Introduction**

Mango is one of the important 6tropical fruits of the world. Mangoes are a popular, nutritional tropical fruits, which are now one of the most important fruits crops in tropical and subtropical areas of the world. They originated in India, were they have been cultivated for more than 4000 years. Beginning in the 16th century, mangoes were gradually distributed from India to other tropical countries in Asia such as the Philippines, Indonesia, China and Thailand. They were also spread to the Americas in the 18th century, and from western Mexico they were carried to Hawaii in the early 19th century. Now a day, mangoes are grown for commercial and nutritional purpose in orchards and home gardens.

**Mango**: (*Mangifera indica* L) belonging to family Anacardiaceae is the most important commercially grown fruit crop of the country. It is called the king of fruits. India has the richest collection of mango cultivars. In southern Asia, especially eastern India, Burma and the Andaman Islands, the mango has been cultivated, praised and even revered in its homeland since ancient times. Buddhist monks are believed to have taken the mango on voyages to Malaya and Eastern Asia in the 4th and 5th centuries B.C. In 1833, Dr. Henry Perrine shipped seedling mango plants from Yucatan to Cape Sable at the southern tip of mainland Florida but these died after he was killed by Indians. Seeds were imported into Miami from the West Indies by a Dr. Fletcher in 1862. From these, two trees grew to large size and one was still fruiting in 1910 and is believed to have been the parent of the ‘no. 11’ which was commonly planted for many years. The mango became one of the most familiar domesticated trees in dooryards or in small or large commercial planting throughout the humid and semi-aid lowlands of the tropical world and in certain areas of the near-tropics such as the Mediterranean area, Egypt, southern Africa and southern Florida. There is no record of the introduction of the mango into south Africa but a plantation was set out in Durban about 1860.
Materials and Methods
Physical Studies
Studies on different solid media for viability and maintenance of *Pestalotiopsis mangiferae*
Five different solid media viz. Potato Dextrose Agar, Nutrient Agar, Cornmeal Agar, Sabourauds Dextrose Agar and Czapek’s Dox Agar were tested. The best medium was used for further maintenance, multiplication and selection of suitable medium for physiological studies.

Different solid media used
Different solid media and their composition used during the course of present investigation are given below.

**Solid media**

- **a) Potato Dextrose Agar (PDA) Medium**
  - Peeled potato slices: 200 g
  - Dextrose: 20 g
  - Agar: 20 g
  - Distilled water: 1000 ml
  - pH: (6.0)

- **b) Corn meal agar (CMA) medium**
  - Corn meal, infusion from: 50 gm
  - Agar: 15 gm
  - Distilled water: 1000 ml
  - pH: (6.0)

- **c) Sabouraud dextrose agar (SDA) medium**
  - Mixture of peptic digest of animal tissues & pancreatic digest of casein (1:1): 10 gm
  - Dextrose: 40 gm
  - Agar: 15 gm
  - Distilled water: 1000 ml
  - pH: 5.6 ± 0.2

- **d) Nutrient agar medium (NA)**
  - Peptic digest of Animal tissues: 5 gm
  - Sodium chloride: 5 gm
  - Beef extract: 1.5 gm
  - Yeast extract: 1.5 gm
  - Agar: 15 gm
  - Distilled water: 1000 ml
  - pH: 7.4 ± 0.2
  - Temperature: 25 ºC

- **e) Czapek dox agar medium (CDA)**
  - Sucrose: 30 gm
  - Sodium nitrate: 3 gm
  - Potassium phosphate (dibasic): 1 gm
  - Magnesium sulphate: 0.5 gm
  - Potassium chloride: 0.5 gm
  - Ferrous sulphate: 0.01 gm
  - pH: 7.3 ± 0.2
  - Temperature: 25 ºC

Measures of growth
For determining the variation in the colony growth of *Pestalotiopsis mangiferae*, the colony growth of fungus in each petri plate was measured when entire control petri plate was covered by fungus. The colony growth was measured along two diameters at right angles and averaged.

Study of colony and growth of mycelium
Observation for various cultural and morphological characteristics of mycelium viz., mycelia colour, margins, growth pattern, distribution of mycelia growth.

Results and Discussion
The pathogen was grown on five different solid media which were selected for this analysis. Observations on the radial growth rate of *P. mangiferae* in four replications were reported at the end of the 6th day of inoculation. Irrespective of the media used Czapek dox agar (CDA) was found to be the most suitable culture medium for the growth of *Pestalotiopsis mangiferae*. Statistical analysis of the data showed that the radial growth rate of Czapek dox agar (CDA) was found to be statistically higher than that of other cultural media and potato dextrose agar (PDA), sabouraud dextrose agar (SDA) is statistically the same as the growth rate. Whereas Corn meal agar (CMA) medium was found to be a non-preferable culture medium for the growth of *Pestalotiopsis mangiferae*.

Intervals of day 1, 2, 3, 4, 5, 6 CDA was found to be the most suitable culture medium for the growth of *Pestalotiopsis mangiferae*, with a maximum radial growth rate. Similarly, in all intervals SDA was found to be statistically equal to the growth rate of potato dextrose agar (PDA). Where Corn meal agar (CMA) has been found to be a non-preferable culture medium for the growth of *Pestalotiopsis mangiferae*.

| Solid Media | Day 1 | Day 2 | Day 3 | Day 4 | Day 5 | Day 6 |
|-------------|-------|-------|-------|-------|-------|-------|
| PDA         | 1.25  | 2.55  | 3.65  | 4.85  | 6.05  | 6.55  |
| NA          | 1.35  | 2.25  | 3.15  | 4.25  | 5.45  | 6.25  |
| CDA         | 1.65  | 2.95  | 4.05  | 5.25  | 6.45  | 6.95  |
| CMA         | 1.15  | 1.95  | 2.85  | 4.05  | 5.25  | 5.95  |
| SDA         | 1.45  | 2.65  | 3.75  | 4.95  | 5.85  | 6.45  |
| C.D.        | 0.09  | 0.09  | 0.09  | 0.09  | 0.09  | 0.09  |
| SE(m)       | 0.03  | 0.03  | 0.03  | 0.03  | 0.03  | 0.03  |

Table 1: Effects of different solid media on the growth of mycelium of *Pestalotiopsis mangiferae*. 
Discussion
During present studies, five different solid media were tested for culturing of *Pestalotiopsis mangiferae*. Out of which, Czapek dox agar (CDA) was observed maximum mycelia growth followed by saubouard dextrose agar (SDA) which was conformation with the work done by Katherine E. Hopkins (1996) who tested different solid media for culturing of *Pestalotiopsis mangiferae* and concluded that Czapek dox agar (CZA), potato dextrose agar (PDA) medium and saubouard dextrose agar (SDA) supported the best mycelia growth among the solid media. Czapek dox agar (CZA) was found to be the best supporting medium for *P. mangiferae* by Rajeev K Upadhyay & R S Dwivedi (1979); Wahi (1967); Singh (1975) J Chandra Sekhar, et al., (2020) [4] who also used same solid media against the pathogen and similar results were observed.

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Plate 1: Growth of *Pestalotiopsis mangiferae* in different solid media
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