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Mango Black Banded Disease: Exploring Morphological and Cultural Aspects of *Peziotrichum corticolum* (MASSEE) Subramanian

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ABSTRACT: Black banded disease of mango (*Mangifera indica* L.) caused by *Peziotrichum corticolum* (Massee) Subramanian, has become increasingly problematic in recent years, inflicting damage on midribs, twigs, branches, and veins of leaves. This study clearly confirmed the identification of the causative agent as *Peziotrichum corticolum*. The investigation delved into the diverse cultural and morphological characteristics of *P. corticolum* across six distinct media at a consistent room temperature of 27±1ºC, including Potato dextrose agar, Nutrient agar, Czapek's agar, Oatmeal agar, Host extract dextrose agar, and PDA + sugar. Notably, Potato dextrose agar, Oatmeal agar, PDA + sugar, and Czapek's agar emerged as the most supportive media for maximal mycelial growth of *P. corticolum* on the 12th day post-incubation, while Host extract dextrose agar exhibited the least radial growth. Cultures of *P. corticolum* showcased significant diversity in cultural characteristics such as the type of growth, mycelial color, pigmentation, and colony margin.

Keywords: Mango black banded, *Peziotrichum corticolum* (Massee) Subramanian, and Morphological and Cultural characters, Media.

INTRODUCTION

Mango (*Mangifera indica* L.), celebrated as the "King of Fruits" or "Super Fruit", stands out for its robust fragrance, flavor some taste, and rich nutritional profile, making it a prominent horticultural gem in India. Belonging to the Anacardiaceae family, the mango has earned the title of "The First Fruit of India" due to its widespread availability, widespread acceptance, and versatile utility. Recognized for its versatility, mango is a cherished fruit crop extensively cultivated in tropical and sub-tropical regions of Southeast Asia, as highlighted by Vasugi *et al.* (2012). In India, it finds cultivation in states such as Uttar Pradesh, Andhra Pradesh, Karnataka, Bihar, Gujarat, Tamil Nadu, Odisha, West Bengal, Jharkhand, Kerala, and Maharashtra, further solidifying its significance and prevalence in the agricultural landscape.

Mango faces various challenges such as diseases, insect pests, and physiological disorders. In India, major concerns include powdery mildew, anthracnose, mango malformation, and sooty mould or sooty blotch. Among these, the black banded disease, caused by *Peziotrichum corticolum* (Massee) Subramanian, has gained prominence in recent years across all mango-growing regions. Initially considered minor, its severity has escalated. The first report of this disease came from Andhra Pradesh by Reddy *et al.* (1961).

The causal fungus, *Peziotrichum corticolum* (Massee) Subramanian, was initially identified on tree bark in Puna, India, by Massee as documented by Hughes in 1980. Subsequent findings have indicated the presence of this fungus in varying degrees of severity across multiple regions, including Goa, West Bengal, Karnataka, Maharashtra, Bihar, Orissa, Andhra Pradesh, Kerala, Andaman and Nicobar Islands, and Tamil Nadu. Comprehensive studies by Naqvi (2004); Ploetz and Om Prakash (1997), as well as Om Prakash and Srivastava (1987), have contributed to our understanding of the widespread occurrence of this disease.

The disease spreads on tree bark creating big, dark, patchy areas that look like bands that's why it's called that. Fungus also found growing like black velvet on leaf veins and midribs (Pandey and Dinesh 2010); (Gautam *et al.*, 2017).

In recent years, the South Gujarat region has witnessed a conspicuous increase in the prevalence of a specific disease. However, crucial details about the causative fungus remain scarce. In an effort to bridge this knowledge gap, we undertook thorough morphological

and cultural studies to gain comprehensive insights into the nature of the disease in this region.

MATERIALS AND METHODS

The present study on black banded disease of mango was conducted during 2018-2019 at the Department of Plant Pathology, N.M. College of Agriculture, Navsari Agricultural University, Navsari which is situated in South Gujarat Heavy Rainfall Agro-climatic Zone of Gujarat state at 20°95′N latitude, 72°93′E longitude and at an altitude of 9.0 m above mean sea level. The material employed and methodology implemented throughout the investigation are presented here.

Collection of disease sample. The plant disease samples *viz.,* Mango branches, twigs and leaves, showing typical symptoms of the disease were collected from the Agriculture Experimental Station, NAU, Paria as well as from farmer's field.

Isolation of causal organism. We followed the standard tissue isolation procedure to isolate the pathogen from infected bark. The afflicted bark fragments underwent surface sterilization with a 0.1% sodium hypochlorite solution for a duration of 30 seconds and then washed them thoroughly with sterile water. Subsequently, 1-2 sterile bits of infected bark were aseptically transferred onto sterilized Petri plates with Potato Dextrose Agar (PDA) and left them at room temperature (27±1ºC). Plates were monitored at regular intervals for observable growth. Upon the development of fungal growth from the infected tissue, a segment was aseptically transferred to PDA slants and subjected to a 12-day incubation period at 27±1ºC. These slants, now containing a pure culture, were then used for further research.

Purification and maintenance of infecting fungi. The fungal growth of disease causing pathogen obtained from the isolated plant tissue in the PDA media. Further purification was carried out by the single spore isolation technique and maintained the pure culture. Cultures were maintained on PDA slants by sub culturing and stored in refrigerator at 5ºCfor further study.

Sub-culturing occurred on a monthly basis, with these cultures serving as the foundation for our entire study.

Identification of isolated fungi. Fungal growth was critically observed under microscope for cultural and morphological characters. Finally, fungal characteristics observed were compared with the characteristics described in various manuals and by studying morphological characters of the same pathogen as described by previous workers, Reddy *et al.* (1961); Mukherjee and Litz (2009); Patil and Dangat (2012); Gautam *et al*. (2017).

Morphological and cultural studies of associated fungi. In order to study the morphological and cultural characteristics of the pathogen, pure culture of *Peziotrichum corticolum* was grown on different media *viz.,* Potato dextrose agar, Nutrient agar, Czapek's agar, Oat meal agar, Host extract dextrose agar and PDA + sugar.Cultural characters such as the colony diameter, colony colour, type of colony margin and sporulation were recorded.

Spore morphology. The spores were examined by utilizing a 40X magnification with an Olympus CX 31 microscope, the dimensions (length and breadth) of the spores were carefully observed and measured, subsequently leading to the computation of their average size.

RESULTS AND DISCUSSION

Symptomatology. The plant exhibited characteristic symptoms with black banded patterns appearing on midribs, twigs, branches, leaf petioles and leaf veins. Notably, the main trunk was rarely affected. On branches and twigs, distinct black, irregular, velvety fungal growth emerged, expanding in size as the disease progressed. These growths formed large, black, belt-like bands. The velvety texture resulted from the aggregation and erect orientation of fungal mycelium on the bark. Near the infection bands' periphery, young mycelium appeared white or nearly hyaline. Young branches and twigs displayed more prominent symptoms compared to older branches in infected trees. The midribs and veins of leaves exhibit a distinctive black velvety fungal presence, while the petioles often experience construction due to the growth of this dark velvety fungus (Fig. 1).

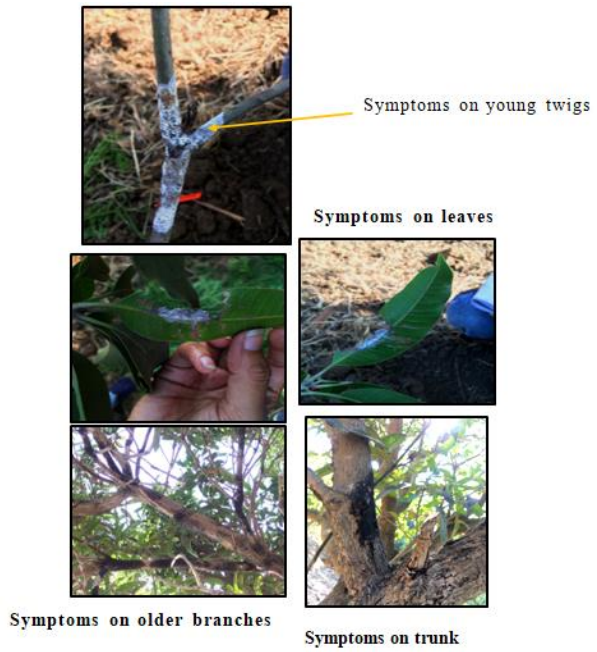

Fig. 1. Symptoms of black banded disease on mango.

Similar descriptions of the symptoms of black banded disease of mango were given by previous workers, Reddy *et al.* (1961); Prakash and Srivastava (1987); Patil and Dangat (2012); Pandey and Dinesh (2010); Gautam *et al.* (2017).

Identification of isolated fungi. The morphological and cultural characters were compared with the description given by earlier workers (Reddy *et al.,* 1961, Mukherjee and Litz 2009; Patil and Dangat 2012; Gautam *et al.,* 2017) and the fungus was identified as *Peziotrichum corticolum*. The fungus in the present study produced initially white or greyish white septate

mycelium which turned into black colour after 10-15 days of incubation.

Morphological and cultural studies of associated fungi. The isolated fungal culture was observed at 40X under Olympus CX 31 microscope to record the morphological details. The microscopic examination revealed that, the septa were formed at irregular intervals. Some cells of the hypha were wider than rest of the cells while some cells were abnormally narrowed at the septum. The mycelium branched profusely in all directions forming a thick tangle of hyphae. On an average, the mycelial width measured 2.95-6.82µm.

Spore morphology. The conidia of fungus were measured and compared with respect to their spore morphology. The conidia were single celled, pale brown, globose, smooth walled, mostly sessile but sometimes with short pedicels, measured 9.50-14.12 µmin diameter (Fig. 2(a)).

Mycelium morphology. Mycelium of *P. corticolum* from infected bark and culture was septate, greyish white to brown in colour and measured average 2.14-5.85µm in width (Fig. 2(b) and (c)).

Cultural characters. Diversity in cultural and morphological characters of *P. corticolum* were studied in six different media at room temperature 27±1ºC as described in "Materials and Methods" and the results obtained are presented in (Table 1 and Fig. 3).

The radial growth, colony characters and sporulation of the fungi were recorded, when the maximum growth was attained on any one of the tested media. The effect of different culture media on the growth of fungi differed significantly. Out of six different media evaluated, Potato dextrose agar, Oat meal agar, PDA + sugar and Czapek's agar media showed Maximum growth of *P. corticolum* mycelium which was 90.00mm. The least radial growth was recorded in Host extract dextrose agar which was 85.00mm. Nutrient agar was found to be the poorest medium for growth of *P. corticolum*, as it showed zero growth of fungal mycelium. Mycelium colour varied from white to light grey. The growth varied from flat to sparse. Pigmentation in the media also varied from brownish yellow to grey.

Further Naqvi (2004) documented the maximum radial growth of *Pestalotiopsis mangiferae*, the causative agent of grey blight in mango on host extract agar.

Table 1: Cultural and morphological characters of *P. corticolum* on different solid media.

Sr. No.	Media	Colony Characters
1.	Potato dextrose agar (PDA)	90mm full gown colony with profuse mycelium which was milky white in colour. Margin was fluffy and light zonation found on back side. Size of spore: 11.75µm. Width of mycelium: 5.85µm
2.	Nutrient agar (NA)	No growth
3.	Czapek's agar	Fluffy greyish mycelium with full coverage *i.e.* 90mm with smooth margin. Centre portion was light and textured. No zonation was found on backside. Size of spore: 9.50µm. Width of mycelium: 2.40µm
4.	Oat meal agar	90mm greyish white mycelium with complete coverage of plate. Mycelium was flat with flat margin. Light three to four zonation found on backside. Size of spore: 9.75µm. Width of mycelium: 3.20µm
5.	Host extract dextrose agar (HEDA)	Greyish white coloured mycelim with cottony growth. Mycelium diameter was 85mm with filamentous margin and inner centre was dark greyish. Strong zonation found on backside. Size of spore: 14.12µm. Width of mycelium: 5.25µm
6.	PDA + sugar	Whitish grey mycelium with 90mm full growth. Mycelium was textured with smooth margin and strong zonation. Size of spore: 11.50µm. Width of mycelium: 4.75µm

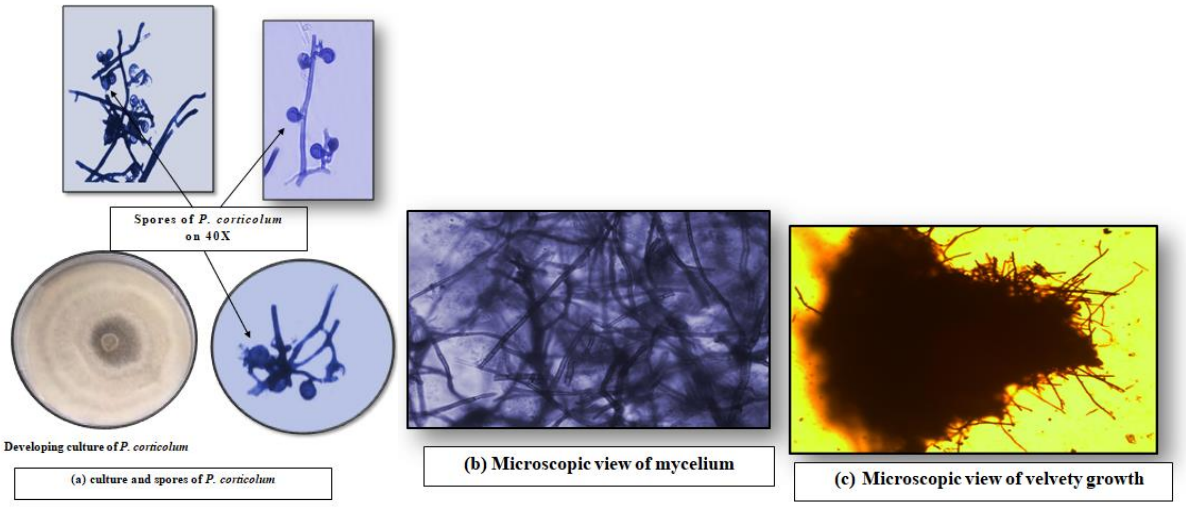

Fig. 2. Morphology and culture of *Peziotrichum corticolum.*

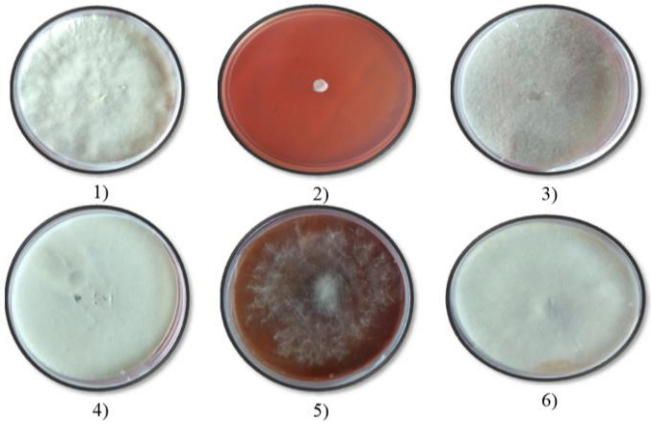

Fig. 3. Growth of *Peziotrichum corticolum* on different solid media after 12 days. 1) Potato dextrose agar (PDA), 2) Nutrient agar (NA), 3) Czapeck's agar, 4) Oat meal agar, 5) Host extract dextrose agar (HEDA), 6) PDA + Sugar.

CONCLUSIONS

In conclusion, this study definitively identifies *Peziotrichum corticolum* as the causative agent of black banded disease in mango trees. Through a thorough investigation of cultural and morphological characteristics on various media, it was determined that Potato dextrose agar, Oatmeal agar, PDA + sugar, and Czapek's agar are the most conducive for maximal mycelial growth of *P. corticolum*. The study highlights significant diversity in cultural traits among different media, including variations in growth type, mycelial color, pigmentation, and colony margin. These findings contribute valuable insights into the biology and optimal growth conditions of *P. corticolum*, providing a foundation for further research and potential management strategies for mitigating the impact of black banded disease on mango crops.

FUTURE SCOPE

The exploration of morphological and cultural aspects of *Peziotrichum corticolum* in the context of Mango Black Banded Disease presents several potential future directions. First and foremost, this study lays the groundwork for in-depth molecular investigations, enabling a more detailed understanding of the pathogen's genetic characteristics and interactions with mango plants. Exploring the pathogenicity mechanisms and host-pathogen interactions at the molecular level could contribute to the development of targeted and sustainable management strategies.

Furthermore, the study provides a foundation for evaluating the efficacy of various control measures, including fungicides, botanicals or biocontrol agents, in mitigating the impact of the disease. Investigating the environmental factors influencing disease development and severity could enhance our ability to implement preventive measures effectively.

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