

Eventually, we documented the first report of *Colletotrichum siamense*-caused leaf spot on *Moringa* (*Moringa oleifera*) in eastern India

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INTRODUCTION

Moringa oleifera common as drumstick tree is grown commonly across a range of tropical and subtropical locations due its nutritional, therapeutic, and economic importance. The tree's leaves, which are of great importance in the nutraceutical and functional food industries, are a rich source of vitamins, amino acids, minerals, and various potent antioxidants. *Moringa* is categorized as a hardy crop. However, in recent years, multiple foliar diseases have been reported in the literature due to increased environmental variability, high-density planting, and continuous monocropping. In the 2021 monsoon season, farmers from Nadia district of West Bengal reported an abnormal leaf spot disease affecting moringa and resulting in a foliage loss of 28-35%. The leaf spot disease had a rapid rate of spread and as such was assumed to be caused by an unknown pathogen. Members of the *Colletotrichum gloeosporioides* species complex have been associated with anthracnose and leaf spot of several tropical crops. *C. siamense*, however, has never been associated with moringa. Reporting new pathogen-host relationships is crucial with respect to forecasting the potential of new disease threats, developing a mitigation framework, and averting possible losses. This short communication is the first to report *Colletotrichum siamense* as the causal agent of the leaf spot disease on moringa.

DESCRIPTION

The Study Materials and Methods Sampling and Isolation: Leaf samples were collected from five commercial moringa orchards (each 0.8–2.5 ha) showing typical symptoms. Symptomatic leaf pieces (5 × 5 mm) were surface sterilized and incubated on PDA at 28 °C. **Morphological Examination:** After 5–7 days colon samples were tested and their characteristics were morphologically, pigmentation, and sporulation. Under the compound microscope (100×) 50 random spores were used to measure the conidia. **Molecular Identification:** A fungal DNA kit was used to extract the genomic DNA. The ITS region was amplified using the primers ITS1/ITS4. For the PCR, the following conditions were used: 94°C for 4 min for the initial denaturation, 35 cycles of 94°C for 45 sec, 55°C for 45 sec, and

72°C for 60 sec, and 72°C for 7 min for the final extension. The sequenced amplicons were aligned and compared to each other using BLASTn. **Pathogenicity Test:** Three-month-old moringa seedlings were kept in a controlled environment chamber. The leaves were sprayed with a conidial suspension (1×10^6 spores mL⁻¹) while the control plants were sprayed with sterile water. For 72 hours to create a humid environment the plants were covered with polythene bags.

Results

Field Symptoms

Symptoms first appeared as small, water-soaked flecks that turned into brown circular spots, 2-5 mm in size. Under sustained moisture, lesions grew to the 8-12 mm size range, centers tan, with dark brown margins. In some instances, yellow halos surrounding center. More serious lesions joined, Extensive debilitating necrosis blight and premature leaf drop coalesced. In severe instances, the 30% loss of infected foliage diminished photosynthetic and moringa leaf rank.

Morphological Features

Isolates grown in PDA had the following features: 7 days of growth white to grey, fluffy mycelium orange-pink conidial masses and conidia that were hyaline, straight, non-septate and enlarged at varying level of 13.8-15.6 × 3.0-4.5 µm These traits were consistent with of *Colletotrichum siamense* description.

Molecular Confirmation

The ITS sequence showed near-identical matches (99.2%) with *C. siamense* sequences in GenBank. Placements in phylogenetic trees further confirmed the identification within the *C. gloeosporioides* s. l. complex.

Pathogenicity Symptoms on the inoculated plants appeared after 7–9 days: leaf spots were identical to field ones and further enlarged and coalesced, while control plants remained free of symptoms. Reisolation displayed colony morphology consistent with the original isolate, thus fulfilling Koch's postulates.

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Significance of the Findings

This study reports the first occurrence of *Colletotrichum siamense* on moringa. This is an important crop for nutrition and medicine. This is significant because *C. siamense* is highly adaptable and is known to infect mango, guava, chilli, strawberry, and papaya, illustrating strong cross-host infection potential. Under highly humid conditions, rapid spread of the pathogen is possible, thus moringa plantations in eastern India (80–90% RH during monsoon) can be particularly vulnerable. This can lead to leaf yield loss, thus affecting the nutritional and medicinal economy reliant on moringa leaves. The remaining part of the pathogen description relates to the *C. gloeosporioides* complex, which is notorious for fungicide resistance and for causing latent infections. The identification of this pathogen at this stage allows for timely supervision and adoption of advanced effective preventive diseases management.

CONCLUSION

This study helps confirm that *Colletotrichum siamense* is newly responsible for emerging leaf spot disease for the first time on *Moringa oleifera* in eastern India. The pathogen's rapid

development in size and ability to cause defoliation represents the first time moringa is facing real challenges to its system of cultivation. Further work should study: state and nation-wide disease surveillance evaluation of bio fungicides and other eco-friendly management alternatives defoliation resistance in moringa plant genotypes genome sequencing to determine specialization on moringa as a host epidemiological projection to determine future disease occurrence under climate change as a result of fast altering morbidity. For the nutritional products industry moringa loses diseases and fast detection with effective management is essential to protect from future disease losses.

Author Contribution S.P: Conceptualization, field survey, laboratory experimentation, molecular work, data interpretation, and manuscript writing.

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Galley Proof