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First report of jackfruit decline caused by *Phytophthora* spp. in Bangladesh

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ARTICLE INFORMATION	Abstract
Article History	A survey was conducted at Gazipur, one of the major jackfruit producing
Submitted: 01 Jul 2020	districts in Bangladesh where decline symptomps of jackfruit trees were
Accepted: 07 Jul 2020	observed. The characteristic symptoms of jackfruit decline included trunk
First online: 16 Aug 2020	canker, wilting, dieback of the canopy and in some cases complete death. The disease incidence was increased with the increase of plants age. Among the
Academic Editor	isolated fungi from the study area, only <i>Phytophthora</i> spp. were found to be
Mohammad Shahjahan Monjil smonjil@yahoo.com	pathogenic and caused decline symptoms in artificially inoculated jackfruit seedlings. Based on the morphological data, the isolates were confirmed as <i>Phytophthora</i> spp. To the best of our knowledge this is the first report of
*Corresponding Author	<i>Phytophthora</i> spp. causing the decline of jackfruit trees in Bangladesh.
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	Keywords: Die back, wilting, canker, sporangium

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1 Introduction

The jackfruit (*Artocarpus heterophyllus*) is called as a national fruit of Bangladesh which belongs to the family Moraceae. It is the second most important fruit tree in Bangladesh after mango. Major jackfruit producing countries in the world are India, Myanmar, Sri Lanka, Malaysia, Indonesia, Thailand, the Philippines, Africa, Brazil, Suriname, the Caribbean islands, Florida, and Australia (Borines et al., 2013). Jackfruit is called as a poor man's food in Bangladesh because of its cheap price but, it is the rich source of calories. Jackfruit production in Bangladesh is 10,31,000 metric tons from 27,334 acres of land which accounts for 7.37% area of the total fruit producing land and 21.42% of the total fruit production in Bangladesh (BBS, 2018). Its average yield in Bangladesh

is 9 t ha^{-1} but, each fruit is weigh up to 35 kg which is the largest known edible fruit (weight up to 35 kg) (Prakash et al., 2009). Moreover, this fruit tree provides timber, fuel, fodder, medicinal and industrial products. However, the production of jackfruit is impeded by some biotic factors. Among these, commonly occurring diseases are leaf spot (Gloeosporium sp/ Phyllosticta sp.), die back (Lasiodiplodia theobromae), fruit soft rot (Rhizopus artocarpi), root rot (Pythium splendens, Phytophthora sp., Fusarium sp., Rhizoctonia sp.); decline disease (Phytophthora palmivora) (Borines et al., 2013); anthracnose (Colletotrichum gloeosporioides) Srivastava and Mehra (2004); corynespora leaf spot (Corynespora cassiicola) (Sangchote et al., 2003). Meah and Khan (1987) recorded 10 diseases of jackfruit in Bangladesh. However, detailed characterization of pathogens is lacking in Bangladesh.

A recent survey at Gazipur district, Bangladesh found that a good number of jackfruit trees above 10 years old were suffered from unidentified decline with symptoms of wilting, defoliation, trunk canker and tree death. Jackfruit decline (*Phytophthora palmivora*) was previous reported in the Philippines (Borines et al., 2013) and Vietnam (Tri et al., 2015). In Bangladesh, the cause of jackfruit decline is yet to be known. Studies on disease diagnosis and causal agents are critical for developing disease management strategies. Therefore, this study was conducted to understand the symptomology of the decline disease, causal agents and their characteristic features, pathogenicity and incidence of this disease.

2 Materials and Methods

2.1 Sample collection

In order to determine the range and extent of decline symptom, a total of 60 plants were randomly selected from Bangabandhu Sheikh Mujibur Rahman Agriculture University (BSMRAU campus) and nearby areas at Gazipur district. The age, disease symptom and incidence of disease (% of trees showing decline symptom) were recorded. Soil samples were collected from the infected areas for the isolation of pathogen. These samples were kept at the refrigerator (4 °C) for isolation of the pathogen.

2.2 Isolation of the pathogen

Soil baiting technique was used following the methods of Callaghan et al. (2016). Rose petals were used as baits with two petals floated on each cup (250 mL) filled with sterilized distilled water. The baits were checked daily for the presence of brown lesions caused by *Phytophthora* which then removed, washed in sterile distilled water then dipped in 70% ethanol for 10 sec then washed in sterile distilled water for one time followed by damp-dried on sterile paper tissues. The infected petals were then placed aseptically onto onion agar (200 g red onion, 17 g agar L^{-1} distilled water) for isolation of the pathogen. A negative control consisted of cup (250 mL) with the same quantity of water and a rose petal without soil.

2.3 Identification and morphological characterization of the pathogen

The morphological characterization was performed from isolated cultures of 7 days old using a compound microscope. Besides, small pieces of agar with mycelia were suspended in sterile water in sterilized petri-plates to induce sporulation which was observed under stereomicroscope (Stemi 508, South Korea) and a compound microscope (Zeiss Primo Star, Carl Zeiss Ltd., Germany). Microscopic photographs were taken using the camera (Axiocam, Carl Zeiss Ltd., Germany) set on top of the microscope using Zen software (Carl Zeiss Ltd., Germany)to observe the hyphae and reproductive structures. A total of 3 isolates were examined for the purpose of morphological characterization.

2.4 Pathogenicity test

The pathogenicity test was performed following the procedures of Borines et al. (2013). Pure cultures of each isolate grown on onion agar for 3-4 days at 25 °C were used for inoculation of healthy jackfruit seedlings. An agar plug (mycelial side down) was placed at the base of seedling (at the soil line) by making a V-cut halfway up the stem. The plug was sealed with parafilm which were removed after 2 days. A total of 5 seedlings in 5 separate pots were used where each pot (30 cm) was filled with 5 kg potting mix (1:1 w/w garden soil and cow dung, sterilized at an autoclave at 115 kPa for 1 h); 4 seedlings were inoculated with pure culture on an isolate but only one control plant was used which was inoculated with mycelial plug without pathogen. These seedlings were monitored daily to observe the symptoms development. After 15 days, the pathogen was re-isolated from soils by baiting with sterilized rose petal and then plating on to onion agar. The isolated cultures were compared with the culture originally isolated from the infected site of sample collection.

3 **Results and Discussion**

The decline symptom was characterized by die back starting from tip of the plant which progressed downward (Fig. 1a), canker at the collar region and trunk of the infected plant (Fig. 1c). Similar decline symptoms were observed by Borines et al. (2013) in the Philippines. The severity of die back was estimated as range between 30-100% in the study areas. The incidence of decline was varied with the age of the plant. In many cases, complete dead of entire plant was observed (Fig. 1b). The death rate was increased with the increase of plant age. The lowest (5%) and highest (30%) death was observed in age limit 10-15 and above 21 years of old plant, respectively. While, the highest incidence (50%) was observed in >21 years old plants (Table 1). Similarly, Borines et al. (2013) reported that older jackfruit plants are more susceptible to decline symptom. From these results, it was expected that the disease incidence would be greater once the sampling size would increase with using older plants (>21 years).

While several organisms were isolated from the soil baiting but *Phytophthora* spp. were consistently found and pathogenic on jackfruit seedling. The *Phy*-

tophthora spp. were identified based on their morphological characters described by Borines et al. (2013). The papillate lemon shaped sporangia (Fig. 2) developed in both soil bait (rose petal) and onion agar are the major morphological characteristics of isolates. All the isolates were showing similar characteristics. Therefore, they were named as *Phytophthora* spp. All three isolates of *Phytophthora* spp. infected



Figure 1. Symptoms of Jackfruit decline (a) partial die back observed in an infected plant, (b) complete death of infected plant, and (c) canker at the collar region and trunk of infected plant

jackfruit seedlings showing complete death 15 days after inoculation. The re-isolation of Phytophthora spp. confirmed Koch's postulates. Other organisms such as Fusarium, Pythium and Colletotrichum were although isolated but they were not pathogenic to the jackfruit seedlings. Thus, these three organisms might be acted as opportunists to damaged tissues. The isolates of *Phytophthora* spp. were pathogenic and isolated only from the soil therefore, better management could be undertaken by amending soils of the jackfruit orchard using proper fungicides at least once in a year. Although, the survey was not representing the whole jackfruit growing areas in Bangladesh but, our results confirmed that *Phytophthora* spp. could cause serious harm to the jackfruit orchard if left untreated. Future studies could be undertaken to increase the survey area and sample size. Moreover, molecular studies are also needed to validate our results.

 Table 1. Disease incidence at different age levels of jackfruit plant

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Age	Ν	Decline symptom (%)	Mortality (%)
10-15	20	30%	5%
16-20	20	40%	20%
>21	20	50%	30%

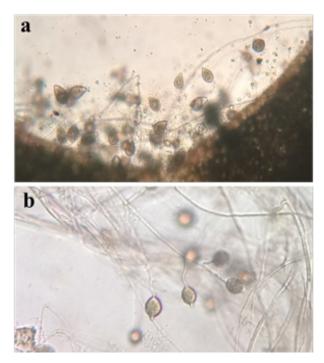


Figure 2. Morphological characteristics of isolates of *Phytophthora* spp., (a) Lemon shaped sporangia were developed from rose petal and floated on water, (b) Papillate, lemon shaped sporangia developed on onion agar at 25 °C for 5 days

4 Conclusions

The decline of jackfruit was characterized by symptoms such as trunk canker, wilting, dieback of the canopy and in some cases complete death. The death rate was higher in older plants. Based on the morphological characters, the pathogen was identified as *Phytophthora* spp. This is the first report of jackfruit decline in Bangladesh.

Conflict of Interest

The authors declare that there is no conflict of interests regarding the publication of this paper.

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