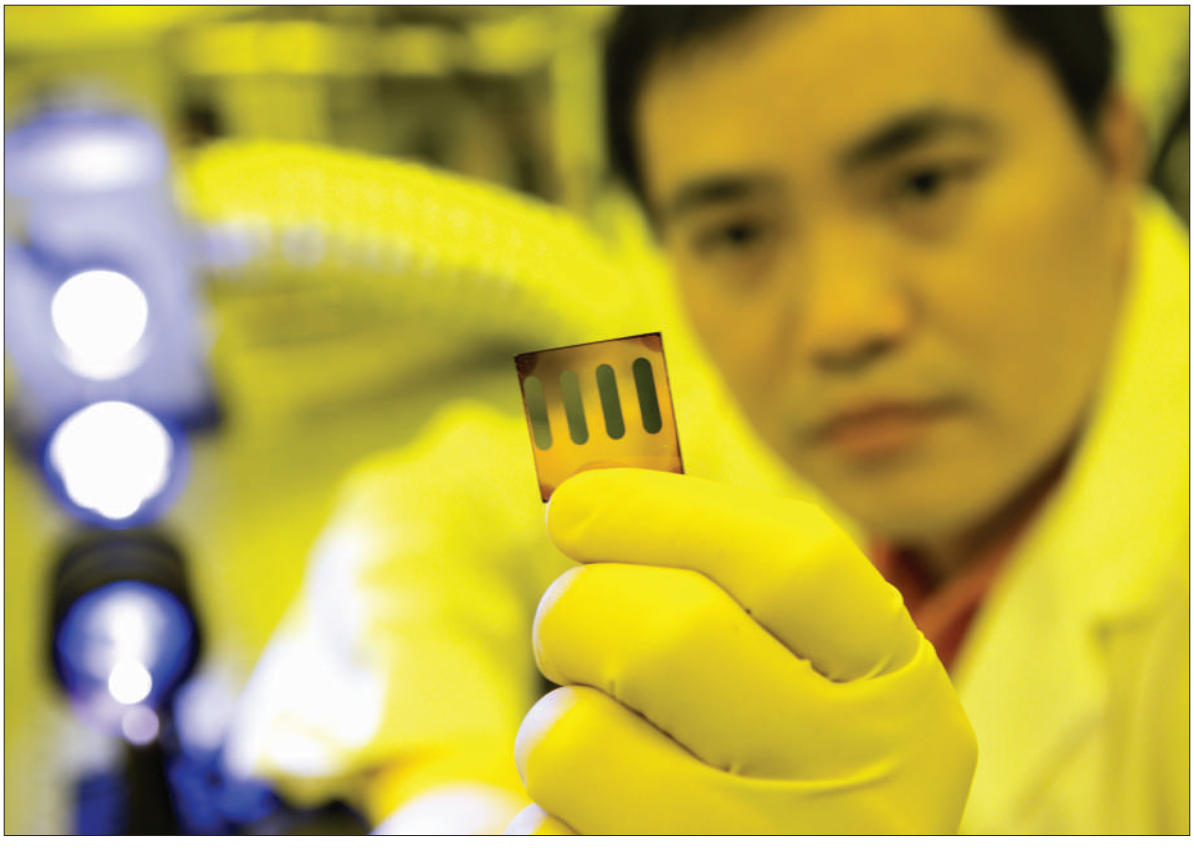

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Research Article
Survey of Leaf Fungal Disease on Urban Tree at Taman Putra Perdana, Putrajaya, Malaysia

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Abstract

Background and Objective: Putrajaya serving as an important administration centre of Malaysia is a local attraction owing to its greenness and a number of recreational parks. Nevertheless, in comparison to roadside trees, there is little or no survey has been carried out on the health status of the tree in these urban parks. Therefore, present study was aimed at a survey on the tree that was infected by leaf fungal disease was conducted in Taman Putra Perdana, Putrajaya to obtain a clear picture of the tree's health status in the area. **Materials and Methods:** The infected leaves of the trees were collected. The fungi that attacked the tree were cultivated in potato dextrose agar (PDA) and agar bacteriological prior for identification. **Results:** From the result obtained, there were 14 tree species at Taman Putra Perdana, Putrajaya were found infected by leaf fungal diseases. Identification of fungi was focused on the 4 tree species with the highest frequencies of infestation. Three fungi species, *Alternaria* sp., *Fusarium* sp. and *Penicillium* sp., were found on the infested leaves of *Michelia alba*, *Lagerstroemia speciosa*, *Shorea multiflora* and *Hopea odorata*. **Conclusion:** Trees at Taman Putra Perdana was suffered from different extent of infestation by a variety of fungi. This study could be useful to site manager responsible for managing the urban trees and preventing severe urban trees loss due to leaf fungus infectious disease.

Key words: Fungus, Putrajaya, recreational park, survey, urban tree

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Urban trees are trees planted in urban area often as ornamental plant. Nevertheless, according to studies done by various researchers, tree at urban area has far more benefits than just serving as ornamental tree[1-3]. Tree can act as wind breaker to deflect and reduce air movement which subsequently prevent soil erosion[4]. Trees also reduce solar radiation[5] by more than 90% and temperature[6] by as much as 5 °C. In a seminal study of how trees might influence the rate of storm water runoff in an urban area, Sanders[7] found that the city's tree canopy cover (22%) reduced runoff by 7%. Moreover, trees are associated with the reduction in both sound and air pollutions[8,9]. Last but not least, serving as ornamental tree, urban trees provide psychological and social benefits. Ulrich[10] reported that the benefits of visual encounters with trees and other vegetation may be the greatest for individuals experiencing stress or anxiety.

However, urban trees are susceptible to various disease attacks. Disease on urban trees will reduce the benefits and to a serious extent, cause plant death. Disease in a plant consists of a series of harmful physiological processes caused by continuous irritation of the plant by primary agent. If the primary agent, the pathogen, is transmissible from diseased plants, the agent is said to be infectious and the disease caused is infectious disease[11]. Viruses, bacteria, fungi and nematodes are far the most numerous and important infectious agent of plant disease[11]. Due to its abundance of green vegetation and recreational parks, Putrajaya serves as an important administration centre of Malaysia and also the attraction for local and foreign visitors[12]. Yet, little or no disease survey targeting on the leaf fungus has been carried out at the area. Such information would be useful for site manager and could be able to prevent infestation outbreak. Thus, the study aimed to carry out a survey on the tree species that was infected by leaf fungal disease in one of the many parks in Putrajaya, Taman Putra Perdana. The fungi that infected the trees was also identified.

MATERIALS AND METHODS

Study area: The sampling location was focused at Taman Putra Perdana park, which is situated at Jalan Putra Perdana, Precinct 1, 62000 Putrajaya, Malaysia. The park is located at the highest point in Precinct 1 and surrounded by Persiaran Sultan Salahuddin Abdul Aziz Shah. It is spread over 70 ha. It acts as an intermediate between the natural domains to the north and the Government and commercial domains to the south and east. The sampling was conducted for 3 months, starting from 27 September, 2017 to 31 December, 2017.

Preliminary survey: A preliminary survey was carried out to determine which trees were infected. Every tree in the park was examined to determine whether it had been infected with leaf fungal disease. All shrubs, palms, vines and creepers are excluded. Trees higher than the heights of the pole used were also excluded. Furthermore, only leaf fungal diseases were taken into account. Diseases caused by non-infectious agents such as lack of essential nutrients are excluded. The species of the infected trees were identified and recorded down along with the signs and symptoms. For this stage, trees with undistinguishable signs and symptoms whether caused by the stated infectious agents or non-infectious agents were also recorded down. The infected part will be collected at next stage for close examination. Photographs and health leaf of the infected trees were taken as a guide to ease the identification of the trees.

Samples collection and sorting: After the preliminary survey was accomplished, samples collection was carried out. The sampling was carried out based on the procedures stated in Jensen and Meilby[13] with some modifications. For this stage, the scope of location in park was narrowed down as information on the infected tree had been obtained beforehand. Areas with the infected tree species were examined. The infected tree was re-identified to serves as a double check for the infected trees. This was crucial to eliminate the possibilities of false identification of the infected tree species. This is also important to know the characteristics of the healthy tree before describing its abnormality. The infected parts of the trees were collected. The samples were then being sorted at lab based on the infected sign and symptoms. The samples were first sort out by two general categories, which are sample with visible fruiting structure and vegetative structure on the surface and samples without those structures on the surface. For samples with fungal infections that showed visible fruiting body on the leaf surface, the samples were examined directly under microscope to differentiate whether they were caused by the same fungus or not. Sample with those visible structures were considered as fungus and are then cultured on potato dextrose agar (PDA). For samples without those structures, they were probably infected by whether fungus, bacteria or virus. Those samples were cultured on both PDA and agar bacteriological. The samples of infected parts were stored in refrigerator to keep it fresh prior to cultivation.

Cultivation of pure cultures and identification: The cultivation of pure cultures was conducted according to Vanangamudi *et al.*[14]. Solid nutrient media potato dextrose agar (PDA) and agar bacteriological were prepared before

samples collection at field. The PDA and agar bacteriological were weighed for 3.9 g, respectively, for every 100 mL of distilled water and poured into a petri dish. Part of plant leave (1×1 cm from the margin of the lesion) that had been infected was cut down and immersed in 10% clorox (sodium hypochlorite) for 1 min. This step was done to increase the chance of obtaining a section free from contaminants. Sections were then removed from the solution with sterile forceps and put in petri dish with distilled water to wash away the clorox. The cut sections were then blotted dry on sterile tissue paper to remove excessive clorox. The infected leave parts were then being transferred to Petri dish that contains solid nutrient media by using sterile forceps and stored upside down in incubator for 4 days. After 4 days, segment of the pathogen growth from leave sections were cut out by using sterile knife and put it on new petri dish with PDA. Pure culture of the pathogen was obtained by sub-culturing a segment of the pathogen growth from previous plate into a new petri dish that contains nutrient media. The sub cultures were kept in incubator until the fungus is fully grown. For identification, semi permanent mounts were prepared. First, a drop of lactophenol cotton blue (LCB) was placed on a glass slide. Then, a portion of the sample was added to be examined and was teased apart with two inoculation needles. The slides were dried for several weeks and were sealed by ringing the edges of the cover glass with fingernail polish. The slides were then used for identification purposes under microscope. Light microscope at 100X magnifications with 10X objective was used.

RESULTS

Species of infected trees: Fourteen tree species at Taman Putra Perdana, Putrajaya were found infected by leaf fungal diseases. Table 1 shows the list of tree species infected with leaf fungal disease and its symptoms. According to the survey carried out, there were mainly two categories of symptoms were observed, which were leaf spot and leaf scorch (Fig. 1-4). Among these 14 tree species, four tree species, *Michelia alba*, *Lagerstroemia speciosa*, *Shorea multiflora* and *Hopea odorata*, were found to be the heaviest infected tree species by leaf fungal disease, with a high number of infected trees recorded. Therefore, the identification on the infected fungi was focused on these 4 tree species.

Identification of fungus that infected the trees: Three species of fungi, which are *Alternaria* sp., *Fusarium* sp. and *Penicillium* sp., were successfully identified from the collected samples of the aforementioned tree species. *Alternaria* sp., a major plant pathogen, was found on the infected leaves of *Michelia alba* and *Shorea multiflora* (Fig. 5, 6). Another 3 fungi species were also found on both the infected leaves of *Michelia alba* and *Shorea multiflora*. *Lagerstroemia speciose* was found infested by *Fusarium* sp., a large genus of filamentous fungi widely distributed in soil and in association with plants (Fig. 7). While the fungus that found on *Hopea odorata* was *Penicillium* sp. (Fig. 8).

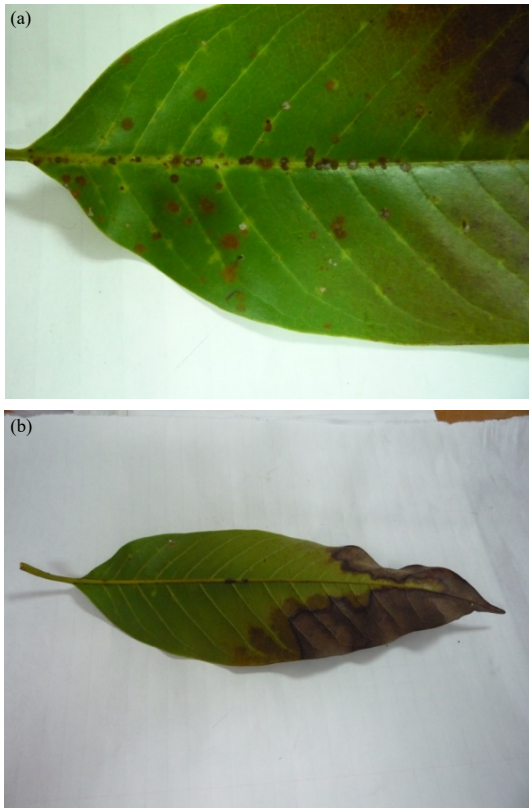

Fig. 1(a-b): Leaf spot and leaf scorch on (a) Upper surface and (b) Under surface of *Michelia alba*

Table1: List of tree species infected with leaf fungal disease and its symptoms

Tree species	Symptoms
Erythryna glauca	Leaf spot
Cinnamomum iners	Leaf spot, scorch
Sara cathaipingensis	Scorch
Michelia alba	Leaf spot, scorch
Alstonia angustiloba	Leaf spot, spot hole
Lagerstroemia speciosa	Leaf spot
Mesua ferrea	Yellowing, spot hole
Syzygium campanulatum	Scorch
Garcinia artrovirdis	Leaf spot
Shorea multiflora	Leaf spot
Calophyllum inophyllum	Scorch
Xanthostemon chrysanthus	Leaf spot
Hopea odarata	Leaf spot
Dryobalanops aromatica	Leaf spot

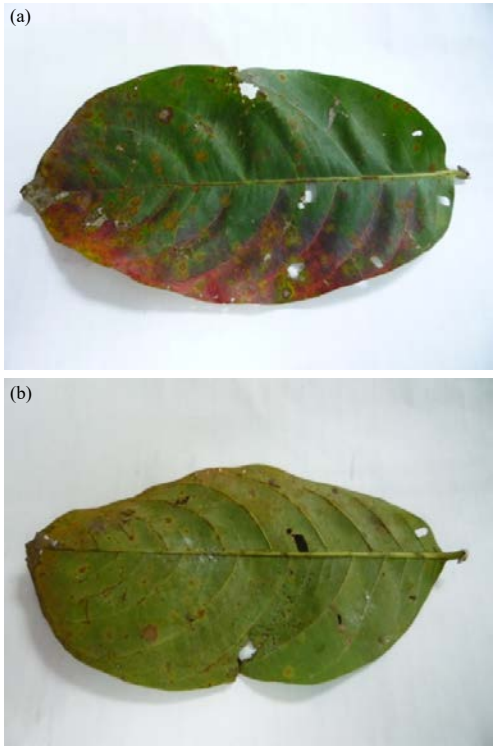

Fig. 2(a-b): Leaf spot and leaf scorch on (a) Upper surface and (b) Under surface of *Lagerstroemia speciosa*

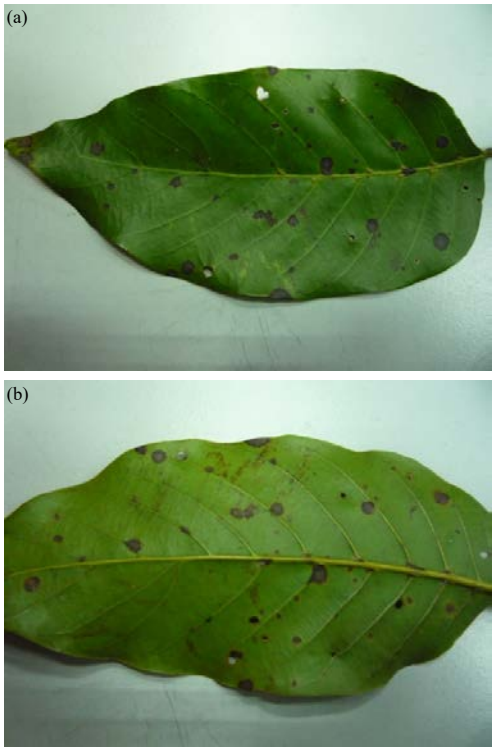

Fig. 4(a-b): Leaf spot on (a) Upper surface and (b) Under surface of *Hopea odarata*

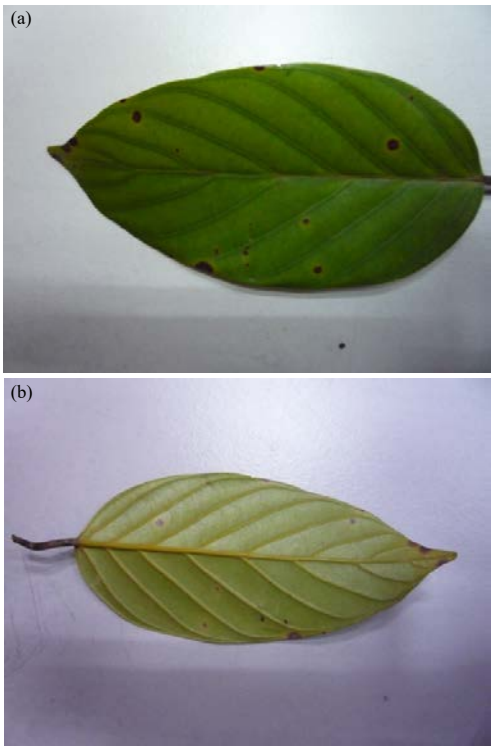

Fig. 3(a-b): Leaf spot on (a) Upper surface and (b) Under surface of *Shorea multiflora*

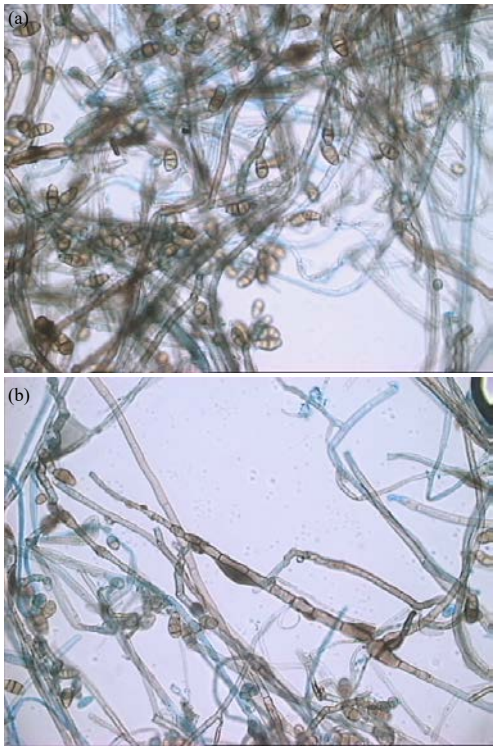

Fig. 5(a-b): (a-b) 40X microscopic view of *Alternaria* sp. found on the leaf of *Michelia alba*

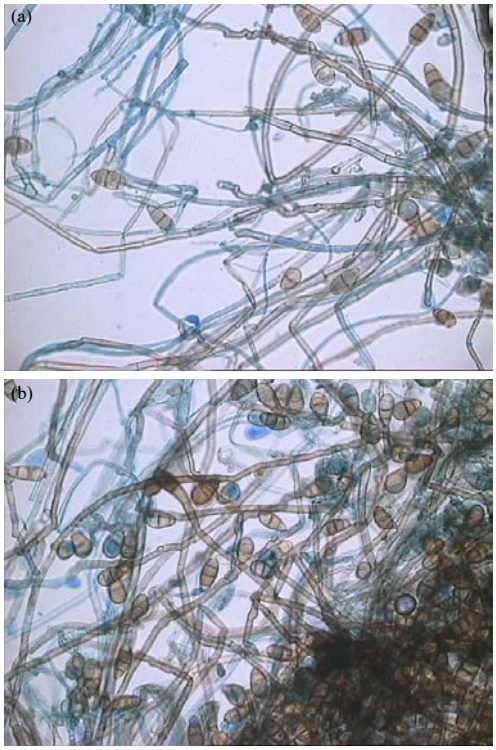

Fig. 6(a-b): (a-b) 40X microscopic view of *Alternaria* sp. found on the leaf of *Shorea multiflora*

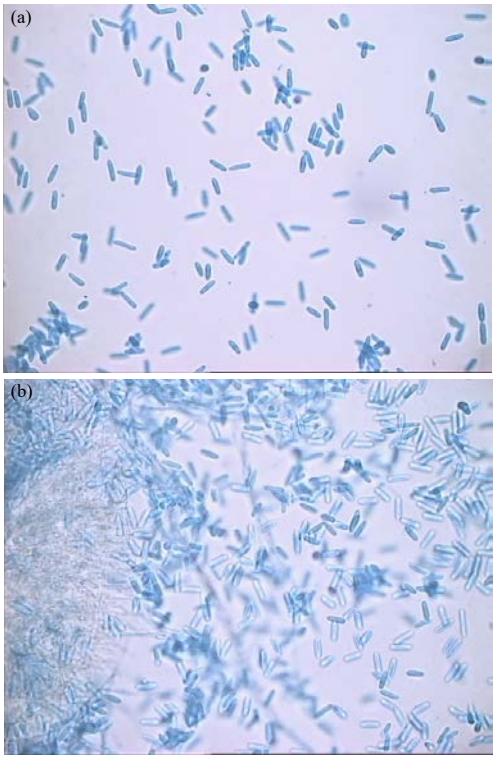

Fig. 7(a-b): (a-b) 40X microscopic view of *Fusarium* sp. found on the leaf of *Lagerstroemia speciosa*

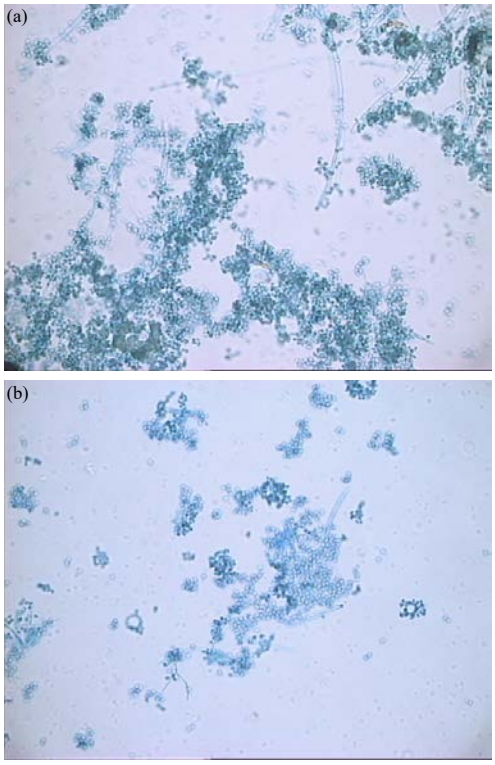

Fig. 8(a-b): (a-b) 40X microscopic view of *Penicillium* sp. found on the leaf of *Hopea odorata*

DISCUSSION

Among the three identified fungi species, *Alternaria* sp. was found dominantly on the infected leaves of *Michelia alba* and *Shorea multiflora*. Due to its dark brown, obclavate to obpyriform catenulate conidia with longitudinal and transverse septa, the fungus that attack *Michelia alba* and *Shorea multiflora* was identified as *Alternaria* sp.[15]. *Alternaria* sp. was known to cause leaf spot and blight symptoms on the leaves of the host plants[16]. Infection of *Shorea robusta* was reported to have caused by a wide variety of fungi, namely *Alternaria* sp., *Fusarium* sp. and *Penicillium* sp.[17]. Infection of these fungi on *Shorea robusta* was found to have adversely affected the viability of the seeds. However, in this study, *Alternaria* sp., was found to be the most dominant fungus species on *Shorea multiflora*. In the case of *Michelia alba*, *Phomopsis* fungi, particularly, *Phomopsis micheliae*, was reported as one of the common fungi that attack *Michelia*[18]. Nevertheless, this fungus was not found on the surveyed trees of *Michelia alba*.

On the other hand, *Lagerstroemia speciose* was found infested by *Fusarium* sp., *Fusarium* sp. is a fungus that resulted in leaf spot of the infected plants[19]. The dominant fungus that infected *Lagerstroemia speciose* was identified

as *Fusarium* sp. This finding was in line with the finding that reported by Ubaid and Suresh[20], who found the leaves of the *Lagerstroemia speciose* was infected by *Fusarium oxysporum*. *Fusarium oxysporum* was also detected on Cavendish banana in India[21]. Meanwhile, the most dominant fungus observed on *Hopea odorata* was *Penicillium* sp., *Penicillium* sp., generally caused leaf blight on the host plants[22]. Previous study reported that *Penicillium* sp., is an endophytic fungus that normally found on mangrove trees[23]. These fungi also reportedly existed in the foliage and sapwood of rubber trees[24]. However, record on the infestation of *Penicillium* sp., on *Hopea odorata* have yet been reported. Based on the survey, it can be observed that the trees at Taman Putra Perdana was suffered from different extent of infestation by a variety of fungi. The results obtained for this study could be useful to site manager responsible for managing the urban trees and preventing severe urban trees loss due to leaf fungus infectious disease. Furthermore, the results of this study might provide better information on the leaf fungal diseases on urban trees at Taman Putra Perdana, Putrajaya for further research.

CONCLUSION

There were fourteen species of trees that found to be infected by leaf fungal disease at Taman Putra Perdana, Putrajaya. The fungi successfully identified were *Alternaria* sp., *Fusarium* sp. and *Penicillium* sp. *Alternaria* sp., was found on the infected leaf of *Micheliaalba* and *Shorea multiflora*. On the other hand, *Fusarium* sp., was found on the infected leaf of *Lagerstroemia speciosa*. For the leaf of *Hopea odorata*, the fungus found was *Penicillium* sp. This study was only focused on leaf fungal diseases of urban trees. Thus, further study can be carried out on other parts of the trees. Future study should also cover other causal agents of infectious disease despite fungus.

SIGNIFICANCE STATEMENT

This study assesses the health status of the trees and discovers the types of fungi that infected the trees at Taman Putra Perdana, Putrajaya. The findings could provide a better information on the severity of the infestation to the site manager for taking potential prevention measure. This study will also serves as a baseline data for the researchers in monitoring the health status of the trees in the park from time to time.

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