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SHORT COMMUNICATION

The host preference of a *Ganoderma lucidum* strain for three tree species of Fabaceae family; *Cassia nodosa*, *Cassia fistula* and *Delonix regia*

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Abstract: Root and butt rot of *Cassia nodosa, Cassia fistula* and *Delonix regia* is caused by *Ganoderma lucidum*. *Ganoderma* root and butt rot is a lethal disease on *C. nodosa, C. fistula,* and *D. regia* trees, and *G. lucidum* was identified as the causal organism of this disease. Susceptibility of these host trees was higher when infected at a young age. All infected trees regardless of the age died after 6-24 months from the appearance of the first sign of the disease. Of these three susceptible host species, *C. fistula* was the most susceptible host, dying six months after the first visible sign of the disease. Host preference of *G. lucidum* was demonstrated when new healthy *C. nodosa, C. fistula* or *D. regia* trees were infected while bypassing other tree species including species of Fabaceae family, which were growing in the close vicinity of the infected trees.

Keywords: *Ganoderma lucidum,* landscape trees, root and butt rot.

INTRODUCTION

The genus *Ganoderma* includes several wood decaying fungi on living trees as well as dead trunks and stumps, and has been recorded mostly in tropical and temperate countries. Generally, *Ganoderma* spp. cause extensive heart rots of standing trees by growing in the central, non-living woody tissues. Several studies have been carried out on *Ganoderma* diseases focusing on economical damage, severity of the disease and host range in many regions such as America, Asia, the Middle East and Europe. *Ganoderma lucidum* has been reported as the causal organism of the heart rot disease of several tree species; *Quercus* spp.[1], *Cocos nucifera*[2], *Camellia sinensis*[3], *Prunus persica* [4], *Vitis vinifera*[5]. Moreover, *Ganoderma* diseases have been recorded on 144 hosts in India, and of those *G. lucidum* has been listed on 91 hosts including *Delonix regia* and *Cassia fistula*. However in that study, the true identity of *G. lucidum* was not confirmed[6]. According to previous studies, several *G. lucidum* strains have been identified in the *G. lucidum* complex[7] having different host specificity.

The dying of *C. nodosa, C. fistula and D. regia* trees due to root and butt rot has been observed in the University of Sri Jayewardenepura premises over the past 14 years. The first observation was made on a 10 year old *D. regia* tree located at one end of a row of trees. The dying has spread in a sequential pattern in different locations of the study area and they can be recognised as disease centres. *C. fistula* and *C. nodosa* are important tropical ornamental flowering trees in Sri Lanka, which are native to Southeast Asia. This study was undertaken to examine the spread of root and butt rot disease in a particular study area and identify the causal agent of the disease.

METHODS AND MATERIALS

Study area: This study was carried out in University of Sri Jayewardenepura premises and its close vicinity covering about 16 hectares.

Disease investigation: According to the available information and the knowledge of tree death in the University premises, four major disease centres were identified for detailed study. Field records and observations were made on signs and symptoms of the disease, other associated trees and the effects on them, temporal and spatial distribution, and the impact of the disease. Approximately 45 tree species were

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monitored regularly. Detailed studies were carried out in order to identify the causal agent of the disease using morphological, anatomical and cultural characteristics. *Macroscopic and Microscopic studies*: Morphological features of basidiocarps, such as shape, size, thickness and margin of pileus were examined. Furthermore, the colour and the texture of the pileus and stipe were also recorded.

Cultural characteristics: Both the pathogen and infested wood chips were cultured on potato dextrose agar medium. Pieces (4x4x2 mm) of pileus and wood chips (4x4x2 mm) were placed on plates on four opposite sides of the plate at right angles, 1cm away from the edge of the plate. Four plates were maintained for each isolate and incubated at 30±1^{0}C. Colony characteristics, hyphal features and chlamydospores were examined under the microscope.

RESULTS

Impact of the disease on hosts

Out of 45 live tree species studied, *C. nodosa*, *C. fistula* and *D. regia* were infected by *G. lucidum* and basidiocarps were observed on the base of the trees. Two single annual badsidiocarps were observed at the base of the live tree trunk, 5-15 cm above ground level as the first sign of the disease on *C. fistula. C. nodosa* and *D. regia,* 3-5 single fruit bodies were observed on the live trunk, 5-20 cm above ground level. 5-8 fruit bodies on exposed roots within 1 m radius of the base of trees were also observed. The initial symptom, wilting of the leaves appeared after a few months of the formation of basidiocarps followed by yellowing of the leaves, defoliation, and finally dieback was observed in the crown. The time taken to develop symptoms and death varied with species and the age of the hosts (Table 1). Foliar symptoms appeared on *C. fistula* five months after observation of basidiocarps and died one month later. Symptoms appeared on *C. nodosa* and *D. regia* 10-22 months after exhibiting the first sign of the disease and died 1-2 months later. However, relatively old trees died 8-24 months after the appearance of the first sign of the disease.

Excavated roots of the diseased trees showed root-to-root contact with neighbouring trees. The wood of the butt region of dead trees appeared off-white and had lost its hardness compared to sound wood.

Disease centres

Centre 1 was located on the main access road to the university as a row of trees along the road sides. The row comprises *D. regia* and *Tabebuia rosea* with 5m intervals between trees. In addition *Jacaranda mimosifolia*, *Roystonea regia* and *C. nodosa* were also located in the close vicinity. Four *D. regia* trees ages between 11 to 22 years died due to root and butt rot disease within a 14-year period in a sequential pattern beginning at one end.

Centre 2 was a row of *C. nodosa* and *Mangifera indica* trees (at 4 m intervals) along the multi-storey building located on grassland and associated with scattered *Bambusa vulgaris, M. indica* and *Mesua ferrea* trees within 3-8m distances. Three *C. nodosa* trees (ages between 14 to 18.5 years) died along the row over the

Table 1: Occurrence of the disease at different centres

Disease centre	Tree species	Age; first sign of disease appeared. years(y), months(m)	Age; tree died years(y), months(m)	Diseased tree species	Distance from the diseased tree (meters)
1	D. regia	10y	11y	(First discovered)	-
	D. regia	12y 6 m	14y 4m.	D. regia	5
	D. regia	16y	18y 6m	D. regia	5
	D. regia	20y	22y	D. regia	5
	C. nodosa	18y 6m	20y	D. regia	8
2	C .nodosa	13y	14y	First tree in 2nd centre	-
	C. nodosa	15y 3 m	16y	C. nodosa	4
	C. nodosa	17y 3m	18y 6m	C. nodosa	4
3	C. nodosa	15y	16y 6m	First affected tree in 3rd centre	
	C. fistula	16y 6m	17y	C. nodosa	7
4	C. nodosa	15y 4m	17y	First affected tree in 4th centre	
	C. nodosa	18y 4m	20y	C. nodosa	4

study period due to the same disease. *M. indica* trees within the row were not affected by the disease.

Centre 3 was a grass covered courtyard located by the side of a building with scattered *C. nodosa, C. fistula, B. vulgaris, Polyalthia longifolia* and *Leucaena leucocephala* trees spaced around 3-5 m distances between the trees. Two trees, *C. nodosa* and *C. fistula* aged 15 and 16.5 years respectively died of root and butt rot disease. The distance between these two trees was 7m. Another *C. fistula* tree located on the other side (7 m away) of the road was not affected. Many other tree species *L. leucocephala B. vulgaris* and *P. longifolia* were observed in close vicinity of this centre which were not diseased.

Centre 4 was a row of *C. nodosa* and *Pinus caribea* trees along a minor road on the campus. The interval between two trees was about 4 m. Only two *Cassia nodosa* trees 4 m apart died from this disease in the row. In addition there were *Bauhinia variegata, P. longifolia* and *Plumeria rubra* located in the close vicinity, but these tree species were not affected.

Morphological features of the pathogen

Basidiocarps

Few small white button shaped structures appeared singly on the infected trunks, 5-15 cm above the ground level and on exposed roots. Eventually, they elongated into 5-6 cm long, cylindrical to flat, erect immature basidiocarps. It comprised a white top and a reddish-brown glazy stipe. Thereafter, slow growth rate of the basidiocarp was observed and a horizontal hoof-shaped pileus was formed with a stipe attached to the wood. At maturity the pileus was also tightly attached to the wood. The upper surface of the young basidiocarp was glazed yellowish brown with a white margin while the lower surface was white with a glazed reddish brown stipe.

The mature basidiocarp was hoof-shaped, hard, shiny and mahogany-red on top with concentric grooves, and cream to white under surface with a reddish-brown boarder along the pileus margin. The length and width of the mature basidiocarp were 8.0 cm and 9.0 cm respectively. The cross section of the basidiocarp is more or less wedge-shaped while the thickness at the proximal end was 2 cm which thinned out towards the marginal end (0.5 cm). The colour of old basidiocarps changed into dark maroon to black on top and the texture of the pileus changed to be spongy. The underside appeared as ash-brown with a 1.2 cm broad, shiny reddish-brown boarder along the pileus margin.

Hoof-shaped, laterally stipitate basidiocarps were formed singly at the base of the trunk directly attached to the wood. Basidiocarps were observed on exposed lateral roots within 1 m radius of the base of *D. regia* and *C. nodosa* whereas on *C. fistula,* basidiocarps were formed only on the butt region up to 15 cm height of the trunk. The bark where the basidiocarps formed was slightly depressed and cracked.

Cultural and microscopical characteristics

The colony colour, growth characteristics of isolates from pileus and wood chips were more or less the same in this study (Table 2).

The width of dark brown skeletal hyphae of the context was 4.47-7.45µm. Round to elliptical-shaped pores were observed on the underside of the pileus, with pore length of 158-168 µm and width of 141-152 µm. 4-5 pores/mm were observed on the pore layer. Basidiospores were golden brown, ovate, thick-walled and bitunicate. The length and width of spores ranged from 8.94 to 10.43 µm and 5.99 to7.45µm respectively.

DISCUSSION

Macroscopic, microscopic and cultural characteristics of the *Ganoderma* isolates were exactly identical with the characteristic features of *Ganoderma lucidum* complex, which has been described previously by several authors[8,9]. Morphological features of basidiospores of the isolated pathogen were similar to that of *G. lucidum*

Table 2: Features of fungal colonies from pileus and wood chips

Features	Pileus	Wood chip
Colony colour (immature)	Off white	Off white
Colony colour (mature)	Bluish green	Blush to ash green
Chlamydospore length (µm)	51.1-54.9	52.1-55.2
Chlamydospore width (µm)	29.8-32.5	29.3-31.6
Hyphal width (µm)	11.6- 12.6	11.3-12.
Hyphal cell length (µm)	42-70	38-66

strains reported by several authors. Further, the size of basidiospores measured in the current study is comparable with sizes of previous records of *G. lucidum;* 8.5-13x5.5-8.5 µm[10] , 7-13x5-9 µm[11],9-11x6-7 µm[12] 7-13x 6-8 µm[13] and 9.9-11.9x6.2-7.7 µm[14]. The basidispores of *Ganoderma* spp. within the family Ganodermataceae are considered as characteristic taxonomic feature[15]. Hence, the isolated causal organism of root and butt rot of *C. nodosa, C. fistula* and *D. regia* was identified as a *Ganoderma lucidum* strain. Several distinctive strains have been identified in *Ganoderma lucidum* complex from different parts of the world.

Infected trees survive for varying periods of time, depending on the species and the age of the tree. *C. fistula* was the most susceptible tree to *G. lucidum* strain, dying within a relatively short period (6 months) of formation of fruit bodies (signs) while the other two species, *C. nodosa* and *D. regia* showed mild susceptibility, and survived for 1- 2 year periods after exhibiting the first sign. Generally, younger trees of these species are more susceptible to the disease than older trees.

In the current study, the particular strain of *G. lucidum* was identified as the causal agent of only three host species, which include two previously recorded species by Sankaran *et al.*[6] . It is evident that the isolated *G. lucidum* strain shows host preference, and the decay causing pathogen attacks only *C. nodosa, C. fistula and D. regia* trees.

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