

A report on occurrence of *Neofusicoccum* sp. in *Carallia brachiata* (Lour.) Merr. trees from Karnataka, India

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ABSTRACT

Carallia brachiata (Lour.) Merr. is one of the soft woody species occurring all along the Western Ghats region of Kundapura, Karnataka, India. The species is considered to be a potent medicinal plant exclusively used against itch and septic poisoning. A study was undertaken based on the report of forest department (Karnataka) on mortality of *Carallia brachiata*. Observation of symptoms on the infected trees and experimental analysis of the samples under laboratory conditions revealed the incidence of *Neofusicoccum* sp., a first report on *Carallia brachiata*. This particular *Neofusicoccum* sp. belongs to Botryosphaeriaceae family, and is considered to be an important fungal pathogen associated with tree decline in forest and plantation worldwide.

Keywords: Botryosphaeriaceae; Fungal pathogen; *Neofusicoccum* sp.; New species

1. INTRODUCTION

Carallia brachiata (Lour.) Merr. is commonly known as Karalli (Indian medicinal plant) belongs to the family Rhizophoraceae. The tree is an immensely colossal ornamental tree found in semi evergreen forests. Similarly, the species occurs in planes possessing geographical distribution in Malaysia and Australia (Sasidharan 2004). In general, this species consists of medicinal value, where its bark has wound healing and anti- inflammatory activity (Krishnaveni *et al.* 2009, Krishnaveni *et al.* 2009). Not only in recent past times, the use of bark for medicinal purpose like treatment of itch, oral ulcer, inflammation of throat and stomatitis was found traditionally (Nadkarni and Nadkarni 1995). The leaves and bark are additionally used against septic poisoning, while the fruit extracts are important for treatment of ulcer (Krishnaveni *et al.* 2009). The extracts of bark exhibited hepatoprotective activity (Kumari *et al.* 2012) and inhibition of proanthocyanidins namely carallidin, mahuannin A, and para hydroxyl benzoic acid (Phuwapraisirian *et al.* 2006).

The cruds extracts showed antibacterial and antifungal activity (Neeharika *et al.* 2010). Thus, *Carallia brachiata* plays an increasingly important role for its exclusive pharmaceutical applications.

In particular to the state Karnataka, *Carallia brachiata* is found in primary forest and less often secondary forest, freshwater swamp forest and on hills, mostly on peat soil. The long term success of this species, however, may be threatened because of its sudden massive mortality in recent times.

2. MATERIAL AND METHODS

Our survey on mortality of *Carallia brachiata* at Kundapura Forest Division consisted of 3 ranges. The field data were based on observations done from external symptoms of the dead tree, which actually revealed the severity of the tree decline. The dead trees truly disposed symptoms of die back/tree decline. The samples of infected portions of the dead trees were collected for further investigation at laboratory condition. A field survey of diseased trees was conducted in 3 ranges of Kundapura Forest Division, Karnataka (Table 1).

Table 1. Distribution of diseased *Carallia brachiata* trees at Kundapura Forest Division.

S. No.	Ranges	No. of trees surveyed	Partially dead trees	Completely dead trees
1	Kundapura	20	2	18
2	Sanker Narayana	30	5	25
3	Hebri	30	2	28

The infected tree samples were collected in sterile polythene bags and stored in refrigerator for further isolation and identification pathogen. The experiment was carried out at the Plant Pathology Laboratory, Institute of Wood Science & Technology, Bangalore. Infected samples were taken and surface sterilized in sodium hypochlorite solution for 2-5 min and washed thrice with sterile water and dried on sterile blotter sheets and then placed on potato dextrose agar plate medium.

Pure culture were obtained by sub- culturing to PDA medium and maintained on PDA slants at 4 °C. Spores were mounted in lactophenol cotton blue stain and examined under phase contrast microscope. Microscopic observation was recorded and images were captured electronically with a Nikon digital camera.

The pathogens were initially identified based on colony and conidial morphology and compared with previously published reports (Rodas *et al.* 2009, Amponsah *et al.* 2009, Iturritxa *et al.* 2011, Heath *et al.* 2011). Isolates were examined for the formation of conidia. Conidial morphology (shape, cell wall, color, and presence and absence of septa) was recorded using a phase contrast microscope.

3. RESULTS AND DISCUSSION

The results revealed that amongst the 3 ranges surveyed, the diseased incidence ranged from 80 to 90% in all the surveyed forest range. Amongst the dead trees, die back symptom was more prominent in all the three surveyed ranges. The morphological observation from the dead trees consists of reddish brown to black discoloration and lesions on the trunk region (Fig. 1a, 1b). In some cases the bark was removed and exposed wood split open turning to a cavity followed by decay and ultimately die. Other than this, the tree is also affected by certain minor foliar pathogens which were not found to be of most significant.

The laboratory studies revealed isolates of *Neofusicoccum sp.* producing aerial and highly dense mycelium on PDA. Culture were initially white, but becoming grey to black. Based on culture, shape, morphology and optimum temperature for growth isolates had regular colony shape and colour (whitish cream), abundant aerial mycelium and a fast growth rate at 22°C in 48 hours.

Based on the observed morphological/microscopic characters and published report (Rodas *et al.* 2009, Amponsah *et al.* 2009, Iturritxa *et al.* 2011, Heath *et al.* 2011) isolates with hyaline and smooth conidia were identified as *Neofusicoccum sp.* (Fig. 1c).



Figure 1. (a) Infected tree of *Carallia brachiate*, (b) Symptoms showing in bark region of tree, (c) Smooth walled conidia of *Neofusicoccum sp.*

The symptoms or the effect of diseases caused by this fungal species under naturally growing trees is very difficult to judge, however the damage have been found significant, where ever it was observed. More over the most significant facets of Botryosphaeriaceae fungi is some species are found to be aggressive pathogens when trees become stressed.

The countenance of disease for species of Botryosphaeriaceae is virtually completely linked with certain system of stress or non-optimal growth situations of trees (Blodgett & Stanosz 1995; Ma *et al.* 2001; Paoletti *et al.* 2001; Smith *et al.* 1994; Stanosz *et al.* 2001). This was an initial attempt made to study mortality of *Carallia brachiata*, even though the actual cause for the death of the tree was due a complex phenomenon, occurrence of the pathogen *Neofusicoccum sp.* was predominant among the stressed trees. The trees were in-fact very old and naturally growing.

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