

## SHORT COMMUNICATION

# HYPHAL INTERACTIONS BETWEEN *TRICHODERMA VIRIDAE* AND *GANODERMA BONINENSE* PAT., THE CAUSE OF COCONUT ROOT AND BOLE ROT

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**Abstract:** *Ganoderma boninense* Pat. is the causative agent of root and bole rot disease of coconut. Hyphae of *Trichoderma viridae* were always present in the basidiocarp tissue of *G. boninense*. Attempts to separate *T. viridae* from *G. boninense* by culturing basidiocarp tissue on agar media in the laboratory were not successful. Investigations using slide culture techniques clearly showed that *T. viridae* formed coils around the hyphae of *G. boninense* and at the points of coil formation hyphal deformations were observed. These suggest that *T. viridae* may have potential as a bio-control agent against *G. boninense*.

**Key words:** Bole rot, coconut, *Ganoderma boninense*, hyphae, root diseases, *Trichoderma viridae*.

## INTRODUCTION

Root and bole rot of coconut caused by *Ganoderma boninense* Pat. was first reported in Sri Lanka in 1974.<sup>3</sup> It is a serious disease in southern Sri Lanka, the pathogen being very virulent in sandy areas, causing death of palms 6-30 months after infection.<sup>4</sup> Fructifications or basidiocarps of the fungus are formed on the rotted parts of the trunk and on exposed roots. The disease spreads through root contact between adjacent palms and by air borne spores.<sup>3</sup> The soil inhabiting fungus *Trichoderma*, is known to inhibit the growth of several soil borne plant pathogenic fungi.<sup>2</sup> This study reports on antagonism between *Trichoderma* and *G. boninense*.

## METHODS AND MATERIALS

*Isolation of Ganoderma boninense:* Infected coconut palms showing clear symptoms of the disease-rotting on trunks together with basidiocarps, were used to isolate the fungus. *G. boninense* was isolated from tissue obtained from the inner portion of the rotted areas of trunks on malt extract agar [MEA]. Before isolation tissues were surface sterilized with 5% sodium hypochlorite for 10 min. Pure cultures were obtained and maintained on MEA at 30°C.

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*Isolation of T. viridae from basidiocarps:* The basidiocarps of *G. boninense* were obtained from infected coconut palms. In all experiments freshly harvested [less than 24 h] basidiocarps were used. To isolate the fungus from the basidiocarps, pieces of about 1 cm<sup>3</sup> were obtained from different regions of the basidiocarp. The pieces were surface sterilized for 5, 10, 15, 20, 25 or 30 min in either 5% sodium hypochlorite or saturated calcium hypochlorite and washed 3 times in sterile distilled water. Thereafter, the external surface of each piece of basidiocarp was carefully removed by slicing under sterile conditions and the remaining tissue was plated on MEA and the plates were incubated at 30°C.

*Study of fungal interactions:* The slide culture technique described by Wijesundera *et al.*<sup>6</sup> was used. A clean glass slide was placed on a Z shaped glass rod in a 9 cm diameter petri dish and autoclaved. Afterwards, a small amount of sterile molten MEA was poured and evenly spread over the slide to make a thin agar film. One end of the slide was kept free of the medium to facilitate handling. Inocula of *T. viridae*<sup>6</sup> and *G. boninense* [each inoculum a 1 cm<sup>2</sup> agar square obtained from the periphery of 5-d-old cultures on MEA at 30°C] were placed 1 cm apart on the slide. A few ml of sterile distilled water was added to the petri dish to prevent drying and incubated at 30°C for 3 days. Observations were made at 10x40 magnification under a light microscope.

## RESULTS

The cultures resulting from basidiocarp tissue always had both *G. boninense* and *Trichoderma* growing together. Repeated attempts to obtain a pure culture of *G. boninense* from this mixed culture failed. When the basidiocarp tissue was transferred to MEA *G. boninense* grew out from the tissue followed very soon by *Trichoderma*. The growth of *G. boninense* was always restricted only to a small area around the basidiocarp tissue by *Trichoderma*. Soon afterwards, *Trichoderma* grew over the entire *G. boninense* colony. The *Trichoderma* associated with *G. boninense* basidiocarp was identified as *T. viridae*. The identification was based on morphological and reproductive characteristics of the fungus.<sup>5</sup> The slide cultures showed that *T. viridae* formed coils around hyphae of *G. boninense*. Coil formation first appeared 1 d after incubation and increased rapidly upto day 3. At the points of coil formation, the hyphae of *G. boninense* had deformations - the walls were swollen and the appearance suggested wall dissolution. After day 3 observations were difficult due to rapid degeneration of *G. boninense* hyphae.

## DISCUSSION

The results of this investigation indicate that *Trichoderma* has antagonistic effects against *G. boninense*. It formed coils around *G. boninense* hyphae and caused wall deformations. Similar effects of *Trichoderma* against other root

pathogens *Rigidoporus lignosus*<sup>6</sup> and *G. lucidum*<sup>1</sup> have been reported. *Trichoderma* is known to produce several cell wall degrading enzymes such as glucanases and chitinases.<sup>2</sup> The wall deformation associated with coil formation is thought to be due to the activity of these enzymes.<sup>2</sup> The inability to separate *G. boninense* from *Trichoderma* using basidiocarp tissue suggests that in nature *Trichoderma* enters the fructifications and interacts very closely with the hyphae of *G. boninense*. The interactions reported in this study, if further developed, could be used as a bio-control agent against *G. boninense*.

## References

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