The role of Pythiacious soil-borne micro-organisms in the tuart decline at Yalgorup

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Pythiacious soil borne micro-organism, encompassing Phytophthora species, include a diverse group of pathogens that have been identified as contributing and inciting significant forest declines throughout the world, including the south-west of Western Australia and the world. Continuing research indicates that Phytophthora species may have significant, yet unclear, roles in forest declines. Improved methods of molecular species identification indicate a greater diversity of species than once evident from strictly morphological identification, and the ongoing evolution and divergence of new species.

Soil bioassays

The presence of soil borne micro-organisms associated with the decline was deciphered through two in vitro soil bioassays assessing the difference in growth of seedlings grown in non-pasteurised soil, containing soil micro-organisms from declining sites, and pasteurised soil from the same sites. An additional in situ soil bioassay has been conducted with tuart seedlings planted adjacent to the fine roots of declining and non-declining trees (Figure 2). Pythiacious organisms associated with declining seedlings will be isolated through direct plating onto selective and non-selective media.

Isolation of Pythiacious pathogens

To isolate relevant Pythiacious pathogens, root and rhizosphere soil was harvested from declining trees, exposed with the Air-Spade® (Figure 1). Organisms were isolated by baiting rhizosphere soil and symptomatic roots onto selective media, and by direct plating of harvested roots from declining trees and tuart seedlings grown adjacent to diseased roots (Figure 2) and in harvested rhizosphere soil grown in an in vitro soil bioassay.

Assessment of root characteristics of declining tuart

Fine root proliferation, health and mycorrhizal association was measured as a component of the air spading. Declining trees had a lower numbers of fine roots and mycorrhizae associated with exposed root systems than healthy trees. Exposed fine roots from declining trees also showed characteristics of callus formation over the root tips, and proliferation of fine roots posterior to the callused tip. This typical root morphology has been observed as a histological response of Avocado roots to Phytophthora cinnamomi infection (Phillips et al. 1987) as the host, walls of the pathogen.
A statistically significant variation was observed between the growth rates of seedlings grown in organic and nutrient layer soils (Figure 3). All seedlings after three months growth exhibited symptoms of nutrient deficiency.

**Injection trial**

A preliminary cross-classified injection trial was conducted assessing the application of phosphite and Medicap MD®: complete nutrient (nitrogen, phosphorous, potassium, iron, manganese and zinc), zinc, iron and insecticide as a control method for managing the tuart decline.

The role of *Pythiacious* pathogens associated with the decline was interpreted through response of the declining trees to the Phosphite application. Phosphite application has been shown to control some *Phytophthora* declines by inducing defence mechanisms in the host at concentrations not directly toxic to the pathogen. The first year of canopy data, indicating the influence of the treatment minus seasonal variation, shows that phosphite applied at 75 g/L induced a clear improvement in canopy condition (Figure 4). A clear decrease in canopy condition was also observed when phosphite was applied at 50 g/L. A clear increase in canopy condition was also observed when phosphite was applied in combination with the complete nutrient and zinc treatment (Figure 5). The validity of all findings is being further verified through continuing trials. An additional large phosphite application trial was conducted to confirm the uptake efficiency and impact of phosphite application at different concentration rates. The first year results from the second phosphite application trial will be available in February 2007.

**Ongoing research**

- Isolation and identification of *Pythiacious* microorganisms from the continuing soil bioassay.
- Assessment of the impact of the different soil treatments on seedling growth on the continuing soil bioassays.
- Assessment of the impact of the phosphite, nutrient and insecticide treatments from the initial preliminary injection trial, and the different phosphite applications from the second phosphite application trial, will be continued for the duration of the project.
- Three pathogenicity trials will be conducted to test the pathogenicity of: *Pythiacious* microorganisms isolated from declining sites and all *Phytophthora* species identified from the south-west of WA.

**References**
