BANKWEST *LANDSCOPE* CONSERVATION VISA CARD TRUST FUND GRANTS 2002-2003

FINAL PROJECT REPORT – July 2004

Identification of the causal organism associated with stem canker disease in the rare and endangered Meelup mallee (*Eucalyptus phylacis*).

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This study was carried out by Peter Scott as an Honours Project at the School of Biological Science and Biotechnology at Murdoch University in 2003. The Grant was used to cover travel and equipment costs for Peter to undertake the study.

Figure 1. *Eucalyptus phylacis* ramet.

Introduction

The presence of a severe canker pathogen in the rare and endangered Meelup mallee, *Eucalyptus phylacis* (Fig 1), population suggests that it may be in a state of decline, due to a complex range of factors. The presence of stem and branch cankers, and stem and branch dieback, indicate pathogenic activity. The problem may be exacerbated in the future due to the lack of genetic diversity of *E. phylacis* as it survives solely in this single clonal population. The destruction of an unknown number of ramets during road construction in the last 50 years has also impacted on the population.

The major factors posing a threat to *E. phylacis* in the foreseeable future include potential human impact and biotic factors such as fungal pathogens, herbivores, as well as climate

changes. A cursory survey of the condition of the plant population shows clearly that most of the stems and branches of all the ramets exhibit some degree of canker damage (Fig 2), as well as damage due to insects and other factors. The object of this study was to isolate fungal pathogens associated with cankers and identify those that may cause disease in *E. phylacis*.

Figure 2. Cankers on *E. phylacis* stems. A, an open canker in bark tissue; B, severe cankering of bark and defect in wood; C, cross section of stem through canker shown in B.

Results

The population was initially surveyed to determine the impact of the canker disease and insect attack on each ramet. The results showed that within the population stem and branch material more than 5 cm in diameter had a greater percentage of total surface area affected by canker damage, insect damage, open bark splitting and healed bark splitting than material less than 5 cm in diameter (Fig. 3).

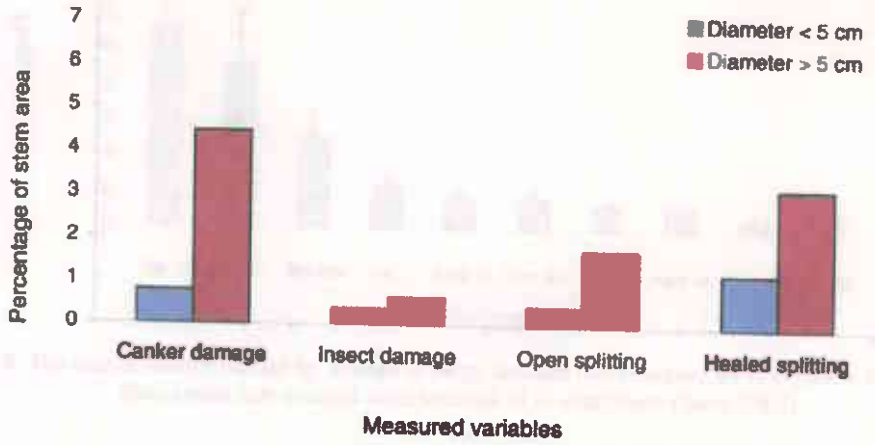

Figure 3. The percent of bark area affected by canker and insect damage within the population of *E. phylacis* (Scott 2003).

Fungal pathogens were the main focus of this study and the main species isolated from the cankers and apparently healthy tissues on the stems and branches of *E. phylacis* included *Botryosphaeria australis, Cytospora eucalypticola, Endothiella* sp. and *Favostroma* sp. The frequency of isolation of both *B. australis* and *C. eucalypticola* indicate that they were both involved in canker formation on *E. phylacis* as well as the surrounding *C. calophylla*. In addition to canker pathogens, *Phytophthora cinnamomi* was isolated from soil within the population. The wide spread susceptibility of *Eucalyptus* spp. to *P. cinnamomi* indicates a significant potential threat that requires further investigation.

Analysis of the genetic diversity of *B australis* isolates obtained from *E. phylacis* ramets, identified a different vegetative compatibility group in each of 19 ramets and a common vegetative compatibility group from 2 ramets. In pathogenicity trials on excised stems of *E. decipiens*, *B. australis* isolates produced significantly larger lesions than *C. eucalypticola, Endothiella* sp., *Favastroma* sp. and several other unidentified isolates (Fig. 4). When a variety of *B. australis* isolates were inoculated into *E. calophylla* and *E. ficifolia* seedlings, they produced a wide range of lesion lengths (Fig. 5). When *B. australis* (Isolate 10b) was inoculated into *E. phylacis* branch tissue, it formed significantly larger lesions that both *P. cinnamomi* and uninoculated controls (Fig. 6). These trials demonstrated that *B. australis* is associated with the development of canker disease in *E. phylacis*, and that the pathogenicity of *B. australis* is variable within the *E. phylacis* population.

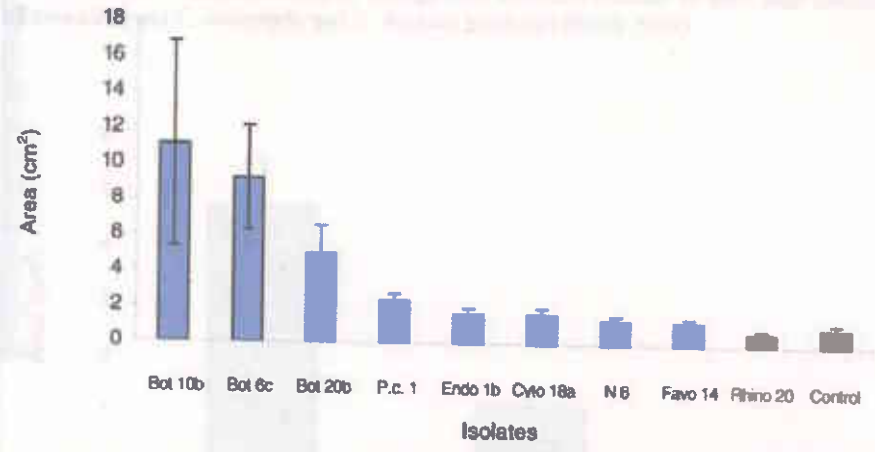

Figure 4. The size of lesions caused by a range of fungi isolated from cankers on *E. phylacis* 20 days after inoculation into excised stem sections of *E. descipiens* (Scott 2003).

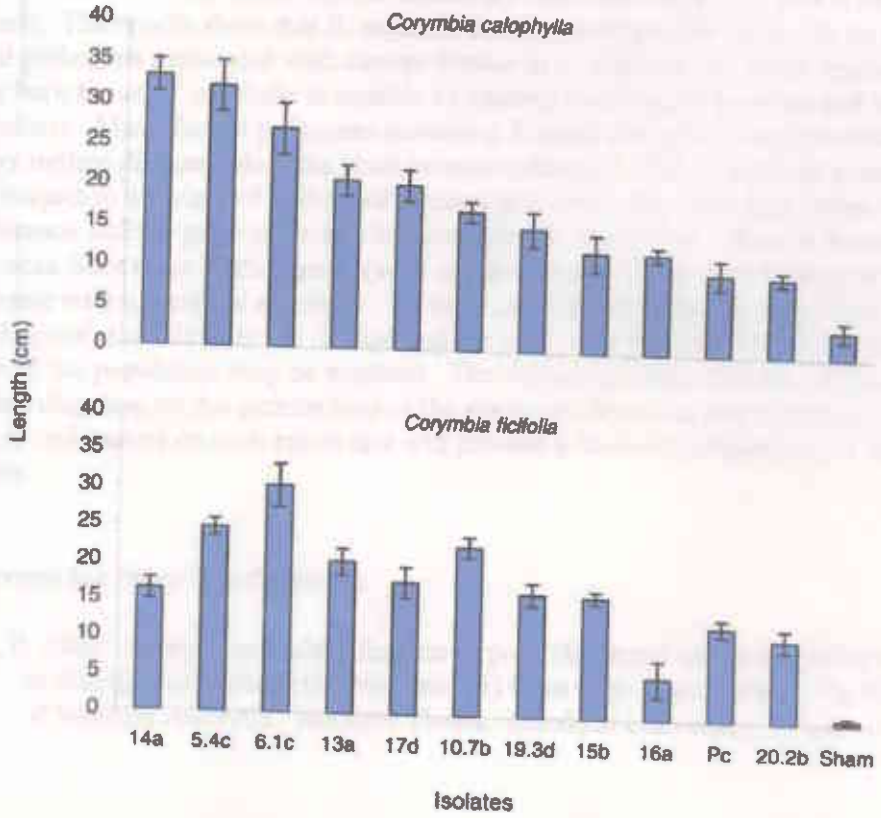

Figure 5. The length of lesions caused by a range of *B. australis* isolates 40 days after inoculation into the stems of 10-month-old *C. calophyla* and *C. ficifolia* seedlings (Scott 2003).

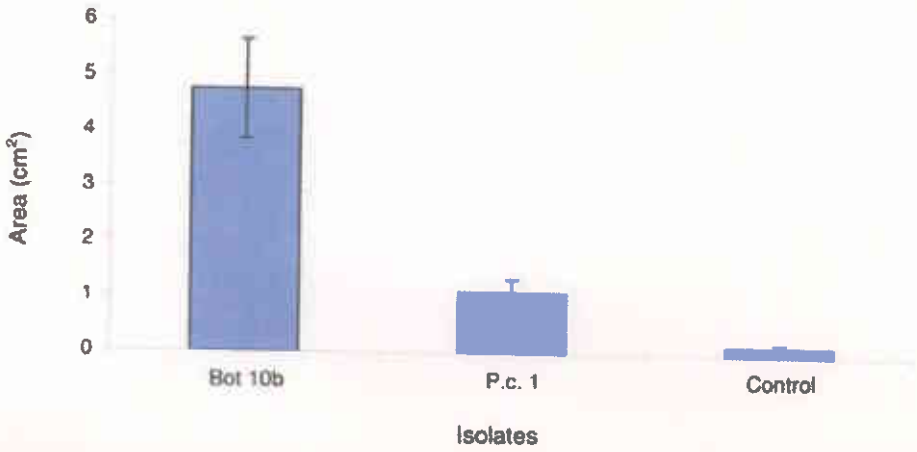

Figure 6. The size of lesions caused by a *B. australis* and *P. cinnamimi* isolates 4 weeks after inoculation into the bark of living *E. phylacis* branches (Scott 2003).

Conclusions

This was the first study undertaken to assess the cause and impact of canker disease on *E. phylacis*. The results show that *B. australis* and *C. eucalypticola* appear to be the main fungal pathogens associated with canker disease in *E. phylacis* and when inoculated into living bark tissue, *B. australis* is capable of causing cankers on the stems and branches of *E. phylacis*. Many fungal pathogens including *B. australis* and *C. eucalypticola* do not always initiate disease unless the plant becomes stressed. The *E. phylacis* population has been subject to a variety of additional stresses in recent times including human interference and the general decline in rainfall in the southwest. These influences and influences from other biotic agents (such as insects) may all be contributing to a disease syndrome with a complex aetiology. To fully understand the relationship between the causal agent(s) involved in the decline and the host, rigorous monitoring of the future health of the population may be required. The project also included the preparation of detailed diagrams on the architecture of the stems and branches and recorded damage by cankers and insects on each ramet and will provide a basis of comparison for future surveys.

Reference for Peter Scott's thesis:

Scott, P. 2003. Analysis and identification of possible causal agents of canker formation in *Eucalyptus phylacis* (Meelup mallee) from Cape Naturaliste in the south west of Western Australia. Honours Thesis. Murdoch University. 154pp.