# Mycorrhizae of the Epacridaceae and Its Use in Propagation

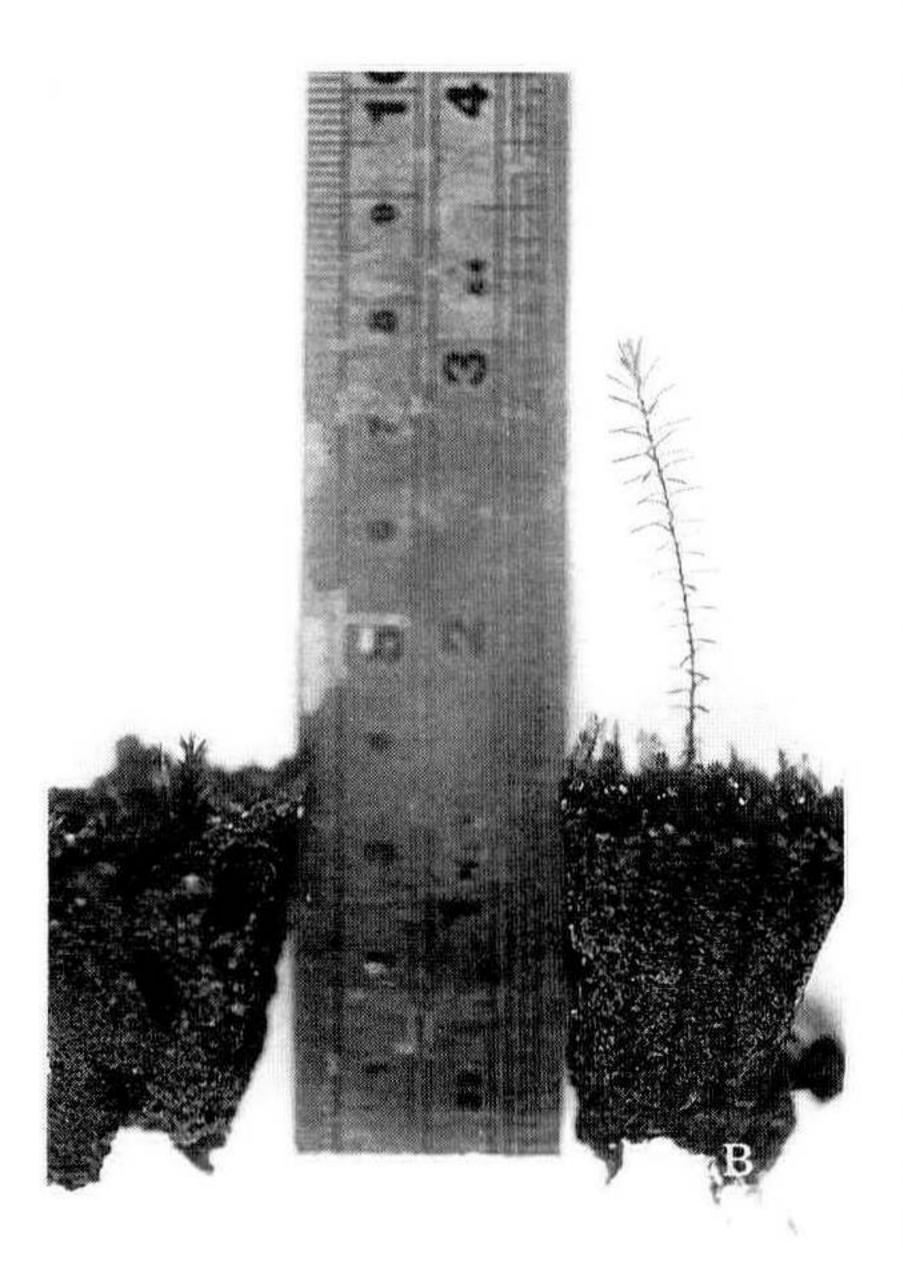
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## INTRODUCTION

Mycorrhizae benefit several commercially produced plants (Galea and Poli, 1994). This paper is a brief overview of mycorrhiza of the Epacridaceae and how they may be used in propagation. Members of the Epacridaceae are difficult to propagate (Thompson, 1986; Williams, 1986). The seeds in some species are extremely small and are not used in propagation, although they appear to germinate readily in the wild (Reed, 1989). Germination rates can be as low as 3% to 7% with *Epacris impressa* Labill. Cuttings are the usual method of propagation; however, success varies enormously—between 0% and 100%. With low survival rates and losses at planting out several attractive epacrids are not grown commercially. This is unfortunate as there is a large public demand for them.

Anecdotal evidence suggested that adding soil from beneath established plants improved the strike rate of epacrids, suggesting that the mycorrhizae which are present in all epacrid roots play a role in establishment. Research was undertaken to investigate the use of mycorrhizae in propagation of *Epacris impressa* and other members of the family. Before continuing with the results of the experiment, it is



**Figure 1.** Six-week-old seedlings of *Epacris impressa* grown in non-mycorrhizal (A) and mycorrhizal (B) soils.

necessary to review the structure of mycorrhizae and discuss how they benefit plants.

# **MYCORRHIZAL FUNGI**

Mycorrhizae are capable of forming a mutually beneficial relationship with the roots of plants (Harley and Smith 1983). Plants that have mycorrhizal relationships are larger and healthier than non-mycorrhizal plants due to increased uptake of water and nutrients, especially nitrogen and phosphorus (Fig. 1). The relationships can be complex, with more than one mycorrhizal fungus inside a plant's roots simultaneously.

Although there are differences in morphology, mycorrhizae can be divided into two broad groups: ectomycorrhizae and endomycorrhizae. Ectomycorrhizal fungi do not penetrate the cortical cells of the root but form a fungal sheath and network of hyphae around the root. This type of relationship is found in woody plants such as Fagaceae, Myrtaceae, and Pinaceae.

In endomycorrhizal relationships the fungus penetrates beyond the epidermis and forms structures in the cortical cells. Several commercially important plants have this type of relationship, such as *Grevillea* spp., *Chamelaucium uncinatum*, and *Impatiens walleriana* (Galea and Poli, 1994). Some plants, such as eucalypts, can have both ectomycorrhizal and endomycorrhizal mycorrhizae simultaneously.

## **ERICOID MYCORRHIZA**

Over the past 30 years there has been extensive research into the identity and culture of ericoid fungi and into the physiological aspects of the relationship in the Ericaceae. Due to similarities in ancestry and structure with Ericaceae, many assumptions have been made about the role of mycorrhizae in the Epacridaceae.

The Epacridaceae and Ericaceae have a similar root structure. These simple roots are described as "hair roots", consisting of a central vascular stele with one or two rows of cortical cells (Harley and Smith, 1983). The mycorrhizal fungus forms a fine weft of hyphae over the root as well as internal structures variously described as arbuscules, coils, or peletons.

Ericoid mycorrhizal fungi can synthesise the plant hormone IAA from the amino acid tryptophan in culture, suggesting a possible role in root initiation in the Ericaceae (Berta and Gianninazzi-Pearson, 1986; Gay and Debaud 1986). This may have implications for nursery production of epacrids.

#### MYCORRHIZAE AND PROPAGATION OF EPACRIDS

Research was undertaken to determine if mycorrhizae could increase the strike rate and improve rooting in *E. impressa* collected from the Royal Botanic Gardens Annexe at Cranbourne, Victoria. As no pure culture of the fungus was available, soil taken from underneath established plants in the wild was added to pasteurised potting mix into which the cuttings were placed. The presence of mycorrhiza improved the strike rate and health of cuttings (Table 1), but the root areas of mycorrhizal and non-mycorrhizal plants were not significantly different (McLean et al., 1994).

**Table 1.** Effect of soil inoculum on survival and development of cuttings of *Epacris impressa*. Figures with the same letters are not significantly different in each column by GLIM analysis using binomial (survival, roots) or Poisson (health, root area) data.

	Survival (%)	Health (%)	Strike rate (%)		Mycorrhizae (% present per cutting)
Inoculum	88a	80a	56a	1.32a	89
No inoculun	n 66b	<b>46</b> b	<b>44</b> b	1.20a	0

(Data from McLean et al., 1994)

Further research investigated whether other epacrid cuttings responded favourably to the addition of soil containing mycorrhizae. Cuttings of *Astroloma pinifolium* R.

Br., A. conostephioides (Sond.) Benth, Brachyloma daphnoides (Sm.) Benth., Styphelia adscendens R. Br, and Epacris impressa from a site in the Grampians were propagated in mix with and without mycorrhizal inoculum.

Increased strike rate and health of cuttings grown in soil containing mycorrhizal inoculum has been observed for *A. pinifolium*, *E. impressa*, and *S. adscendens*. *Astroloma conostephioides* and *S. adscendens* have not shown a significant difference between mycorrhizal and non-mycorrhizal cuttings.

Results from both experiments suggest that the difficulties in propagating these epacrids may be overcome by adding soil containing mycorrhizae. However, using soil as inoculum is not satisfactory for nursery production since it may introduce plant pathogens. It is therefore necessary to produce the fungi in pure culture. If different plants require specific mycorrhizal fungi for growth, inoculation becomes a complicated procedure. However, recent research suggests that this may not be the case (McLean and Lawrie, 1996).

There is likely to be more than one mycorrhizal fungus infecting epacrid roots. At least two morphologically different types of mycorrhizae have been observed in the roots of eight epacrids from three sites. At two of the sites (Cranbourne and Rye) the endophyte had a similar morphology and size (diameter of hyphae). However, at the Grampians' site the morphology of the fungus was different and the hyphae were three times larger. This suggests that the fungi involved in the relationships at the sites are different. Our results are consistent with research in Western Australia (Hutton et al., 1994).

However, this does not necessarily result in specificity of infection. Cuttings of *A. pinifolium* from the Grampians were grown in Cranbourne soil. When the roots were harvested 'Cranbourne-type' mycorrhizae were seen suggesting that there was little host-mycorrhizae specificity. Although several fungi form mycorrhizal relationships with Australian epacrids, it may thus be possible to produce one isolate that can be used in the nursery production of most species.

#### CONCLUSION

Research so far has shown that the addition of soil containing mycorrhizal propagules can improve health and survival of some epacrids. For this information to be of use in routine propagation, the fungi need to be identified and grown in pure culture. Isolation of the fungi is currently being undertaken in a number of laboratories across Australia (Hutton, et. al., 1996; Steinke and Ashfor, 1996).

Once the mycorrhizal fungi are identified and cultured, the next step in this research will be to investigate directly (using pure isolates) the effect of mycorrhizae on the health and survival of cuttings of *A. pinifolium*, *A. conostephioides*, *B. daphnoides*, *E. impressa*, and *Styphelia adscendens* from the Grampians site and *E.impressa* and *Leucopogon ericoides* from Cranbourne.

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### LITERATURE CITED

- **Berta, G.** and **V. Gianninazzi-Pearson.** 1986. Influence of mycorrhizal infection on root development in *Calluna vulgaris* (L.) hull seedlings, p. 674-676. In: V. Gianninazzi-Pearson and S. Gianninazzi (eds.). Mycorrhizae: Physiology and genetics. INRA, Paris.
- Galea, V. J. and R. C. Poli. 1994. The potential for the use of VA Mycorrhizae in nursery crop production. Comb. Proc. Intl. Plant Prop. Soc. 44:52-58.
- **Gay G.** and **J. C. Debaud.** 1986. Preliminary study of IAA synthesis by ericoid endomycorrhizal fungi, p. 678-682. In: V. Gianninazzi-Pearson and S. Gianninazzi (eds.). Mycorrhizae: Physiology and genetics. INRA, Paris.
- Harley, J. L. and S. E. Smith. 1983. Mycorrhizal Symbiosis. Academic Press, London.
- Hutton, B. J., K.W. Dixon, and K. Sivasithamparam. 1994. Ericoid endophytes of Western Australian heaths (Epacridaceae). New Phytol. 127:557-566.
- Hutton, B. J., K.W. Dixon, K. Sivasithamparam, and J. S. Pate. 1996. Diversity of endophytic fungi in Australian Epacridaceae. Ann. Bot. (in press).
- **McLean, C., A.C. Lawrie,** and **K.L. Blazè.** 1994. The effect of soil microflora on the survival of cuttings of *Epacris impressa*. Plant and Soil 166:295-297.
- McLean, C. and A.C. Lawrie. 1996. Site differences among epacrid mycorrhizae Ann. Bot. (in press).
- Reed, M. 1989. Ericoid mycorrhizas of Styphelieae: Intensity of infection and nutrition of the symbionts. Aust. J. Plant Physiol. 16:155-160.
- **Steinke, E.** and **A.E. Ashford.** 1996. Characteristics of fungi associated with the naturally infected mycorrhizal hair roots of *Leucopogon parviflorus*. Ann. Bot. (in press).
- **Thompson, W.K.** 1986. The effects of origin, time of collection, auxins and planting media on rooting of cuttings of *Epacris impressa*. Sci. Hortic. 30:127-134.
- Williams, R. 1986. Research into propagation of Australian native plants. Comb. Proc. Intl. Plant Prop. Soc. 36:183-187.