



DO INDIVIDUAL HOST TREES SUPPORT DIFFERENT COMMUNITIES OF ECTOMYCORRHIZAL FUNGI?

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Fig 1. Typical native Scots pine woodland (wide spacing between trees that have laterally well-developed canopies – dominant ericaceous understorey vegetation)

1. INTRODUCTION

- There are few field studies looking at EcM diversity associated with individual adult host trees of the same species
- So far the only method to link an EcM root tip to a tree has been to physically trace it back to the stem
- To our knowledge there are no previous records of EcM diversity being traced to individual host trees by molecular methods, i.e. targeting plant DNA in EcM root tips
- Native Scots pine populations have high intraspecific genetic variation which is reflected (and measured) by variation in the monoterpene profile (= “chemotype”) of individual pines.
- Monoterpenes also have a functional ecological role in e.g. soil nutrient cycling & herbivory
- Few studies on effects of monoterpenes of host on EcM fungi^{A,B}

2. MAIN HYPOTHESIS AND AIMS

- ➔ Chemotypically different individual host pines support the same EcM communities
- Develop a molecular microsatellite-based (“fingerprinting”) method to identify individual host tree from EcM root tips in addition to identifying the EcM fungus – from a single DNA extraction
- Sample a chemotypically variable area of native Scots pine woodland to test hypothesis

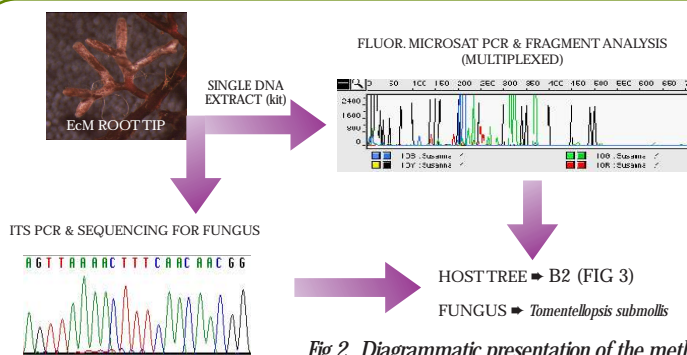


Fig 2. Diagrammatic presentation of the method

3. MICROSATELLITE FINGERPRINTING – METHOD

- Primers for several different pine microsatellite (nuclear) loci were screened using ³²P-labelled PCR, using foliar tissue
- Analysis of pine DNA at three loci (i.e. 3 primer pairs^C) was sufficient to distinguish between 5 individual pines
- Microsatellite analysis was then optimised for detection using fluorescently labelled primers and an automated sequencer - using DNA from individual EcM root tips (Fig 2)

4. FIELD SAMPLING

- An area of native pine woodland (c. 0.7 ha) with small-scale chemotype variation was selected (Fig 3) and sampled
- A total of 177 root fragments from 40 soil samples were morphotyped and EcM root tips analysed further for fungus and host tree ID
- A successful host pine ID was determined for 166 (94%) root fragments from EcM root tips

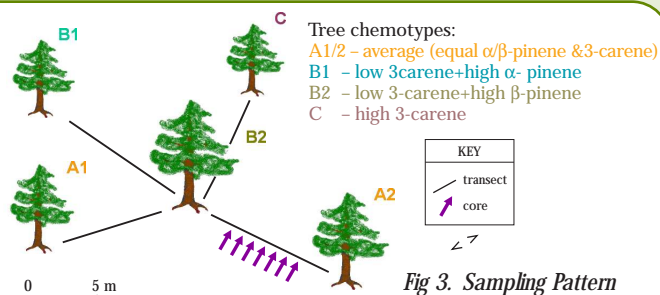


Fig 3. Sampling Pattern

5. CONCLUSIONS

- A new high-through-put method (microsatellite fingerprinting) was developed to identify individual host tree from EcM root tips (using an automated DNA sequencer for data collection)
 - allows screening large numbers of samples
 - microsatellite primers exist for several other EcM host species therefore this method could be applied to other hosts & ecosystems
- Initial results indicate differences in EcM communities between individual trees
- Sampling will be repeated to replicate each tree chemotype and further analyses are still on-going on the EcM fungi