Forest Structure and Fungal Endophytes

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Interactions among endophytic fungi, woody plants and herbivores are usually studied emphasizing fungal mediated plant-herbivore interactions. Seeking evidence for indication of causal relationships between fungi and herbivores sharing the host plant has been self-feeding reasoning in endophyte literature since the findings that some endophytes, particularly seed-borne grass-endophytes in agronomic arena, negatively affect herbivores. Although many studies with natural grass systems, and especially with horizontally by spores transmitted tree-endophytes, have shown more variable effects, alternative approaches have seldom been brought up for discussion. Here, I suggest that the mutualistic nature of plant fungus interactions via enhanced herbivore resistance should be reconsidered especially when focusing on woody plants, and focus on the question, how genetic, spatial and age structure of forest affect the species distribution and frequency of endophytes.

First, we compared phenotypic and genetic frequency correlations for two endophytic fungal genera (*Fusicladium* and *Melanconium*) and birch rust (*Melampsoridium betulinum*) with the performance of six invertebrate herbivores growing on the same half sib progenies of mountain birch (*Betula pubescens* ssp. *czerepanovii*) in two environments over a 3-year period. We found very little support for causal association between fungal frequencies and performance of herbivore species. Results suggest potential causal association only between fungal frequencies and growth performance of one late season herbivore species, *Dineura virididorsata*. Instead, genetic correlations, particularly between autumnal moth (*Epirrita autumnata*) and birch rust, suggest that herbivore performance may be affected by (1) genetic differences in plant quality for fungi and herbivores, or (2) genetic differences in responses to environmental conditions. Later on, genetic analysis (RAMS-PCR) of *Venturia ditricha* (anamorph *Fusicladium betulae*) revealed that (1) host genotypes and environment influence the probability of infection by particular endophyte genotypes, (2) genetic variation correlated negatively with infection frequencies of the fungus, and (3) the susceptibility of the host to a particular endophyte genotype may change when environmental conditions are changed (environment-host genotype interaction). Furthermore, the study on foliar endophyte frequencies in two native (*Betula pendula* and *B. pubescens*) and three exotic (*B. ermanii, B. platyphylla* and *B. resinifera*) birch
species and their hybrids demonstrated that the resistance of hybrids was generally very close to the more resistant parent supporting the hypothesis that the resistance of birch to these fungi is genetically based and caused by dominant inheritance of resistance traits. In short, these results propose that the seemingly direct interactions between herbivores and fungi may actually indicate genetic differences in plant quality for fungi and herbivores or responses to environmental conditions.

We then examined how environmental fragmentation affects the species distribution and frequency of horizontally transmitted endophytic fungi of birch leaves. The Archipelago Sea provided an excellent opportunity to do this. Our study system consisted of 14 islands, ranging in size and distance to the mainland and five mainland study sites. Both *B. pubescens* and *B. pendula* birch, if available, were sampled from these study sites. The leaves were surface sterilized and fungal colonies were grown out from the sampled leaves. The species composition and frequency of endophytic fungi was observed and their occurrence and frequency was compared in relation to e.g. distance to the mainland and land area around the study site. We identified 24 fungal endophytes to species/genus level and detected 12 non-sporulating morphotypes. The frequency of the three commonly detected fungal species, *Fusicladium betulae*, *Gnomonia setacea* and *Melanconium betulinum* were studied in more detail. Our results reveal that frequencies of endophytic fungi in birch leaves depend on e.g. the size of the island, explaining 32-35 % of the total endophyte variation, and distance to the mainland explaining 29-35 % of the variation. According to our study the largest islands near the mainland had the highest foliar endophyte infection frequencies, but the proportions of endophytic fungi vary among commonly detected species.

Finally, we studied effects of silviculture and local environmental variables on endophyte frequencies. We sampled silver birch (*B. pendula*) leaves from seedling stands (5 transects), managed mature forest (7 transects) and natural old forest (5 transects) in eastern Finland. The sapling stands had the highest endophyte and the managed forest the lowest total infection frequency. The old natural forest tended to have the most diverse identified fungal species community, but the difference was not statistically significant. The most frequently isolated endophytic fungi were *Fusicladium betulae*, consisting 70 % of the isolates from the sapling stands and 31 % and 21 % of the isolates from the managed and natural forest, respectively. It is probable that the sapling stands had plenty of available spores combined with otherwise favourable microclimate. In the natural forests *Gnomonia setacea* was the most frequently isolated endophyte genera (30 %), while in the sapling stands only 4 % of the isolates belonged to *G. setacea*. In natural forest *G. setacea* infections were positively correlated with stand age indicating that *G. setacea* is favouring the old forest habitats.

In short, fungal endophytes appear to be affected by all examined factors, genetic, spatial and age structure of the forest. However, we could not detect strong evidence for endophyte-plant mutualism in terms of increased plant resistance to herbivores.
Since many of these fungal endophytes or their close relatives are dormant saprophytes or latent pathogens, recognizing the driving forces of patterns and frequencies of endophytic fungi enables us to understand changes in mycoflora caused by fragmenting previously continuous habitat due to forest practices.

References