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Leaf Litter and Leaf Age as Factors Affecting the Assemblage of Endophytes Associated with Particular Hosts

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Summary

Particular importance has been given to the potential effect of forest canopy on enhancing the rate of initial endophytic fungi colonization of seedlings. Here we compare the rate of endophyte accumulation when endophyte-free seedlings are placed under intact forest canopies with intact or removed leaf litter. The results show a much more rapid accumulation of endophytes in the presence of intact leaf litter. This not only suggests that dead leaves appear to be a primary source of inoculum of foliar endophytic fungi (FEF) for leaves in the understory, it further suggests that local FEF sources (i.e., the local litter) can dominate the composition of colonizing spores. The results of this experiment suggest a mechanism explaining reports of relatively fine scale local differentiation of FEF communities within the same host plant.

Introduction

Extensive surveys across several host plant species (1-8) suggest that different hosts differ in the species composition of their foliar endophytes. Although many fungal endophyte species appear to be present in many, if not all hosts in a given locale, the relative abundance of those endophytic fungi usually differ strikingly among hosts. Specifically, endophyte species that dominate the assemblage in one host often do not dominate in other host species.

Available data from studies in which cohorts of newly flushed leaves are followed through time suggests that the diversity of endophytes usually declines as leaves age (6). One previous study has emphasized the importance of closed (versus open) forest canopy on enhancing the rate of initial endophyte colonization of seedlings (9). However, these experiments confounded the factors, canopy cover and leaf litter. Here we compare the rate of endophyte accumulation when endophyte-free seedlings are placed under intact forest canopies and forest gaps with intact or removed leaf litter.

Material and Methods

The present study was conducted in the Barro Colorado Nature Monument, a tropical moist forest reserve in the Republic of Panama and using *Theobroma cacao* (Malvaceae) as the host plant for the experimental work.

Endophytic fungi free seedlings of *T. cacao* were obtained by sowing seeds in pots containing sterile soil and raising the plants under a plastic chamber which prevent seedlings to be infected from aerial propagules (see 6 and 9). Three treatments (seedlings exposed to different conditions) were compared: 1) intact forest (closed canopy with intact litter; n=15); 2) forest gap (open canopy but with leaf litter intact, n=16); 3) intact forest (closed canopy, with ~90% leaf litter removed within >20 meters of the seedlings, n=16). One leaf from each seedling was sampled after one week of exposure, with 64 pieces of 2-mm^2 per leaf assayed for endophyte infection (assayed as in ref. 9).

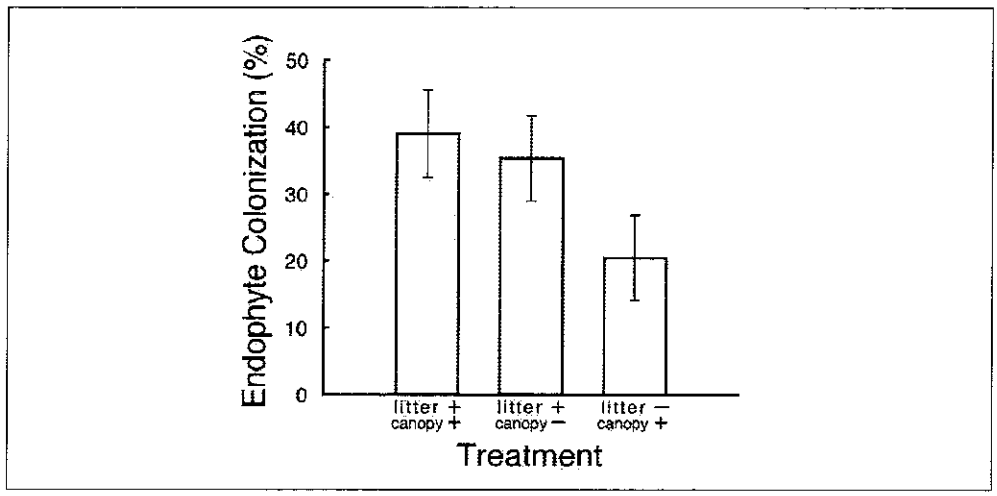

Figure 1. Local leaf litter is a more important source of foliar endophytic fungal inoculum than intact canopy cover. Mean proportion (± SE) of leaf tissue colonized by endophytes in endophyte-free seedlings of Theobroma cacao *after 1-wk exposure to each of three habitats. Kruskal-Wallis One-Way Analysis of Variance, Test Statistic = 7.914,* P = *0.019, 2 df.*

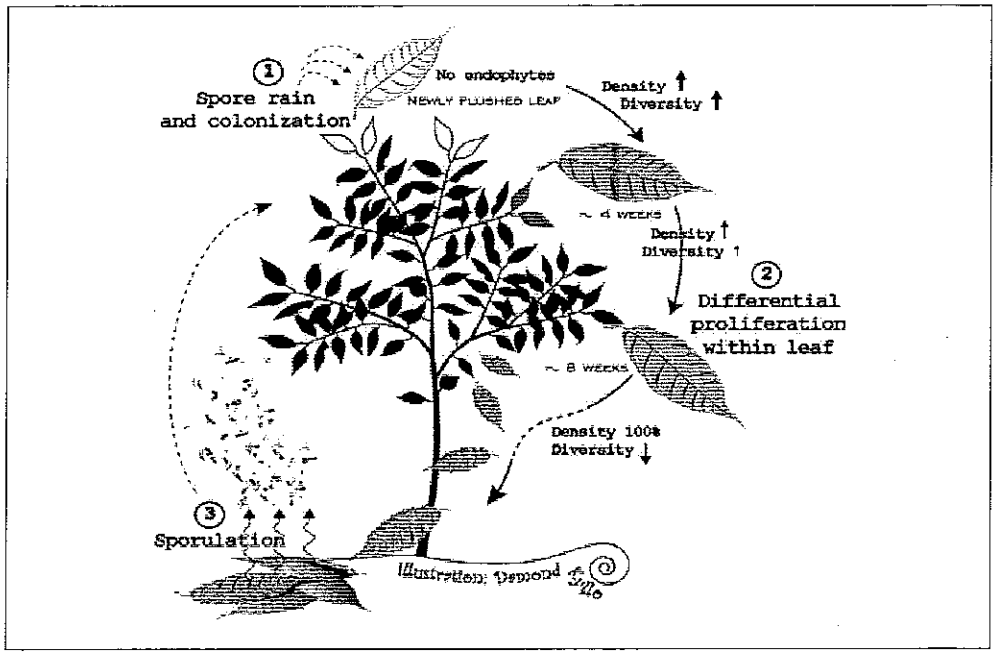

Figure 2. Proposed life cycle for tropical foliar endophytic fungi (FEF) and their host plants. Leaves are flushed, essentially free of FEF, spores land on the leaf surfaces and, upon wetting, germinate, and penetrate the leaf cuticle. After a few weeks, the density of FEF infection within the leaf appears to saturate with a very high FEF diversity. Over several months, FEF diversity usually declines. After leaf senescence and abscission, FEF sporulate and the cycle begins anew.

Results

Colonization of *T. cacao* seedling leaves by endophytic fungi occurred more rapidly in the presence of intact leaf litter (fig1). Leaf litter alone promoted more colonization of leaf seedlings by endophytic fungi than forest canopy (fig 1). These results coupled with available data (6) suggest that leaf litter plays an important role in early colonization of leaf seedlings (see fig 2).

Conclusions

The results show a much more rapid accumulation of endophytes in the presence of intact leaf litter at least for leaves in the understory. This not only suggests that dead leaves appear to be a primary source of inoculum of foliar endophytic fungi (FEF), it further suggests that local foliar endophytic fungi sources (i.e., the local litter) can dominate the composition of colonizing spores. These results may explain potential mechanism behind reports of relatively fine scale local differentiation of FEF communities within the same host plant species.

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