

# Community composition of soilborne pathogens in an old field: Implications for *Prunus serotina* seedling survival and succession

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## ABSTRACT

- The Janzen-Connell Hypothesis suggests that host-specific predators help maintain tree species diversity.
- Soilborne plant pathogens may serve the same function as host-specific predators in the Janzen-Connell Hypothesis because they may depress seedling survival near conspecific trees.
- Previous studies have indicated that *Prunus* seedling mortality decreases with increasing distance from the parental tree and that at least one *Pythium* species plays a direct role in reducing seedling survival close to the parent tree.
- Reverse-dot-blot hybridization (RDBH) methods were used to compare soil *Pythium* community composition near a hedge row populated with *Prunus* trees to 75 meters from the hedge row in a recently abandoned agricultural field.
- No differences in community composition between the hedgerow and field were found in the preliminary data. These similarities provide insight to the spatial patterns of *Prunus* colonization during succession.

## INTRODUCTION



Tagged cherry seedlings beneath a mature cherry tree (above). Seedlings showed higher mortality from damping-off close to the tree (below).



According to the Janzen-Connell Hypothesis, the host-specific plant predator play a large role in the spatial distribution of trees in mature tropical forests by preventing the dominance of any one tree species, thus increasing the overall diversity of the forest (Janzen, 1970, Connell, 1970, Fig. 1). We have applied the Janzen-Connell Hypothesis to our study of temperate forests, using *Pythium* as a pathogen of *Prunus serotina*, black cherry trees.

*Pythium* are soil-borne oomycetes that cause "damping-off disease" in *Prunus serotina*. This pathogen can infect epidermal or cortical cells of the plant, where it releases enzymes that break down pectic cells of the plant, where it releases enzymes that break down pectic plant tissue. Seedlings are highly susceptible to pathogen-induced mortality; older plants are less likely to die as a result of infection because cell wall lignification in mature trees confers protection. Therefore *Pythium* species living on and around roots of mature *Prunus* can lead to distance- and or density-dependent seedling mortality.

Previous studies have indicated that *Prunus serotina* seedlings have a lower survival rate at close proximity to the parent tree, and an increased survival rate far from the parent tree (Packer and Clay 2000, 2003). Though *Pythium* is commonly thought to be a generalist, mortality beneath *Prunus* trees appears to be host-specific (Packer and Clay 2000). A newly described species, *Pythium atrantheridium*, has been isolated from the roots and rhizosphere of *Prunus* seedlings and shown to cause high mortality in *Prunus* seedlings in a controlled greenhouse experiment (Packer 2000).

Based on the above studies, we expect the distribution of *Prunus* trees to be hyperdispersed. However, in young forests this is often not the case. Earlier age *Prunus* communities may not be as limited by *Pythium* pathogens as mature communities because the pathogen has either 1) not yet dispersed to the site, or 2) it has dispersed to the site, but environmental conditions of an early *Prunus* community (high sunlight, decreased soil moisture, etc) allow the plant to escape mortality. In an earlier examination of this question, seedlings were grown in soil collected beneath cherry saplings in an early successional field and beneath mature cherry trees in an old-growth forest. Results indicated that *Pythium* was already established in the early successional field (approx. 25 yrs old) (Fig. 2).

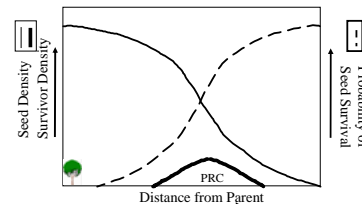
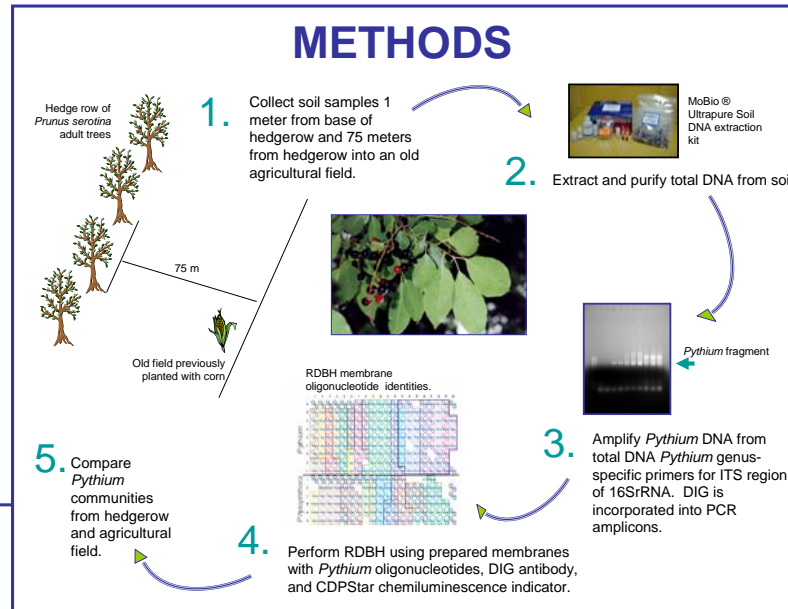


Fig. 1. Graph of the Janzen-Connell Hypothesis. Seed density decreases and seed survival increases at greater distances from the parent tree.

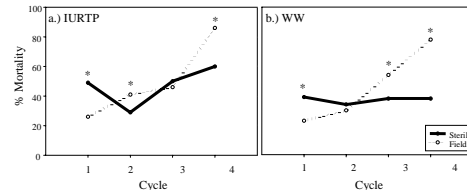


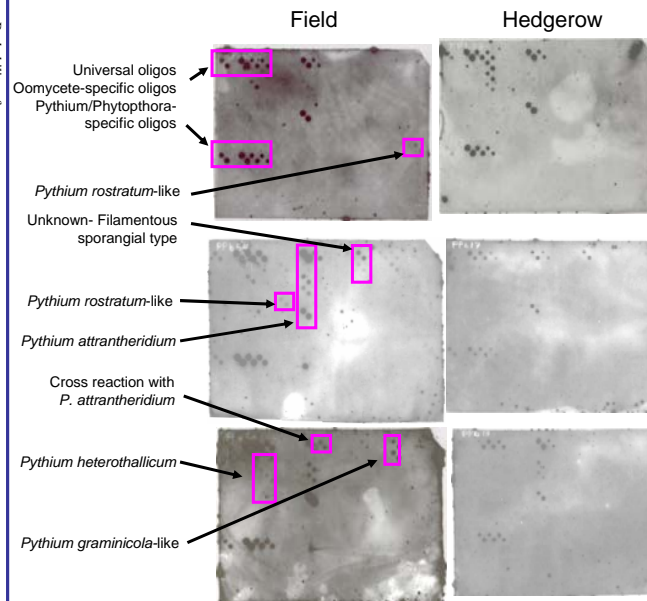
Fig. 2 Effects of sterilization on seedling mortality varies over consecutive growth cycles in soil from beneath cherry trees in a.) an early successional site (~25 yrs) and b.) a mature forest (significant site\*sterilization\*cycle interaction; asterisk indicates  $p < 0.05$  for sterilized vs. field soil comparison). (Packer and Clay 2004)

This study investigates the differences in *Pythium* soil community composition at a hedgerow of mature *Prunus* trees, and 75 meters from the hedgerow in a recently abandoned agricultural field where trees are not yet established. Results from this research provide preliminary data on the establishment of *Pythium* communities during early succession.

## DISCUSSION

- The preliminary data of this experiment indicate that *Pythium* community composition at the hedge row does not significantly differ from the community composition in the recently abandoned agricultural field. Cherry frequently colonizes early during the course of succession. If the pathogen is already present at that point, as these preliminary data suggest, than the environmental conditions characteristic of early successional communities may be allowing cherry seedlings to escape mortality.
- This field could be unusual because it is surrounded by a hedgerow comprised of mature cherry trees. The pathogens found on cherry roots may be absent in a field that is not in close proximity to mature cherry trees. This question will be explored in future studies.
- Though RDBH is a reliable method for determining community composition of known species, it is difficult to use RDBH dot intensity as an indicator of concentration, especially among different membranes. The dot intensity can vary with time of exposure, amount of CDP\*Star used, or concentration of DNA. Also, as with any PCR-based techniques, PCR artifacts may distort concentration of species if the primers have a greater affinity for some template sequences. Because of all of these reasons, RDBH is an unreasonable quantitative measure of species composition. There may be a greater abundance of pathogenic *Pythium* species in our hedgerow samples that cannot be distinguished using these techniques. Future investigations will involve the development of RealTime-PCR techniques to increase quantitative integrity.
- Additionally, RDBH only accounts for previously described *Pythium* species for which distinguishing oligonucleotides are available. For example, *P. atrantheridium*, a recently described species by Allain-Boulé et al. (2004), was isolated from the roots of cherry seedlings in a previous study by Packer and Clay (2000) and subsequently added to the *Pythium* RDBH membrane profile. Therefore, if there are unknown *Pythium* species in the soil, than differences in composition between the hedgerow and the agricultural field may exist that are not detectable in our study. We are currently isolating and sequencing pathogens directly from cherry seedling roots. This will allow us to identify undescribed species not represented on the membrane.

## RESULTS



## Literature Cited

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