Recruitment & Plant-Soil Feedback:

The Effects of Mature Trees on Seedling Survival

By

Samantha Marie Wolf

A thesis submitted

in partial fulfillment of the requirements

for the degree of

Master of Science in Conservation Ecology

(School of Natural Resources & Environment)

at the University of Michigan

April 2013

Thesis Committee:

Assistant Professor Inès Ibáñez, Chair

Professor Donald R. Zak

TABLE OF CONTENTS

ABSTRACT

Plant-soil feedbacks (PSFs) are mechanisms by which plants alter the structure, chemistry, and biology of soil which then influences plant survival. Over time, PSFs have the ability to alter forest composition because the soil biota associated with adult trees differentially affects the establishment of conspecific or heterospecific seedlings. Greenhouse studies have shown that host-preferred pathogens act as a type of PSF that can potentially infect different species of seedlings. We conducted a field study over two summers comparing the survival of four seedling species underneath three adult tree species in a temperate forest. We applied fungicide to the soil surrounding half of the seedlings in order to exclude the most common fungal pathogens. We found that initial stem height and soil moisture significantly influenced seedling survival for all species. Possibly due to unusually extreme drought, we observed great variability in survival rates, and so we found no statistically significant differences between our test and control groups. However, we were able to quantify the effects of the pathogenic fungal community associated with specific tree canopies on the survival of conspecific and heterospecific seedlings. We found indications of conspecific seedling-canopy species interaction which affected survival, although this varied among species. *Prunus serotina* seedlings treated with fungicide experienced a ~92% increase over the average survival of seedlings under control conditions. Comparisons found that under control conditions, all seedling species were negatively affected by the fungal communities associated with *P. serotina* canopies except for *E. umbellata*. Conversely, fungal communities associated with *A. saccharum* canopies tended to have the weakest affect for all native species. Overall, we found that

seedlings treated with fungicide tended to experience increased survival, though the relative degree of effects varied. In summary we did not observe strong changes in PSF effects between treatments of conspecific canopies on seedlings survival but we were able to detect differential survival probabilities due to the fungal community which may contribute to the coexistence of these species.

INTRODUCTION

One of the central questions in plant ecology is how species coexistence is maintained. Basic population ecology posits that two competing species will compete until the superior species drives the other species to extinction (Gause 1932). If both species were equal competitors, then the same scenario of exclusion could occur due to drift (Porter and Gates 1969). However, observations in natural areas often exhibit multiple species coexisting and competing in the same community. Such evidence clearly indicates that more factors are involved in community diversity than simple species competition, factors that promote species coexistence.

In addition to abiotic resource availability and biotic interactions such as competition and mutualism, one type of biotic interaction that also seems to shape recruitment patterns is plant-soil feedbacks (PSFs) (Mills and Bever 1998; Ehrenfeld, Ravi, and Elgersma 2005; Kulmatiski et al. 2008; Bever et al. 2010; McCarthy-Neumann and Ibáñez 2012). Plant-soil feedbacks are mechanisms by which individual plants and entire communities can alter the structure, chemistry, and microbial community of soil via root exudates and leaf litter (Van der Putten et al. 1993; Ehrenfeld et al. 2005; Bever

et al. 2010). These alterations subsequently influence plant survival and growth which is otherwise affected by light, soil moisture, and temperature (Ehrenfeld et al. 2005).

Plant-soil feedbacks may have positive and negative effects occurring simultaneously creating a net effect upon plant survival (Klironomos 2002). Past studies indicate that the negative effects driven by species-specific plant pathogens may have stronger effects upon seedlings establishing under conspecific canopies compared to heterospecific canopies (Mills and Bever 1998; McCarthy-Neumann and Kobe 2010a; Brandt et al. 2013). Therefore, the presence and relative virulence of a species-specific pathogen can facilitate the coexistence of other species that are tolerant of the pathogen (Connell 1971; Van der Putten et al. 1993; Bartelt-Ryser et al. 2005).

This result of negative PSFs via plant pathogens can be represented by the Janzen-Connell (JC) model (Reinhart et al. 2005; McCarthy-Neumann & Kobe 2010a). The JC model (in terms of tree recruitment) describes the likelihood of seedling survival as dependent on the relative proximity of seedlings to a species-specific enemy such as herbivores or pathogens (Packer and Clay 2000; Reinhart & Clay 2009; McCarthy-Neumann & Kobe 2010b). The JC model can be scaled to the adult tree-seedling interaction where species-specific pathogens will be more abundant in soils closer to adult trees (Liu et al. 2012). Seedlings growing near their conspecific parent are more likely to experience greater mortality rates than cohorts establishing far away as greater exposure to species-specific pathogens occur close to the parent (Connell 1971). Recent research has shown, however, that pathogens may vary in species-specificity and target multiple species (Hersh 2009).

3

Seedling death due to negative PSF's is commonly attributed to "damping off" disease facilitated by a variety of soil pathogens. Agents of "damping off" disease include large microbial genera such as *Phytophthora, Rhyzoctonia, Pythium*, and *Fusarium* (Young and Giese 1990). These fungal and oomycete disease agents can include species-specific pathogens as well as generalists that can infect multiple species in a given community (Agrios 1988). Generalist tendencies may appear when the pathogen is exposed to a new potential host (Hersh 2009). Cultures of root-fungal colonies on seedlings grown in soils "cultivated" by another plant species have shown that infections are also possible between heterospecifics (Gilbert and Webb 2007; Hersh 2009). In addition, pathogen species may differentially affect a variety of host-species as different plant species may have more or less tolerance to infection from that particular pathogen (Agrios 1988; Hersh 2009; McCarthy-Neumann and Kobe 2010b). This layer of complexity indicates that seedling recruitment is moderated not only by direct attack from conspecific cultured soil pathogens, but also polyphagic attack from pathogens supported by heterospecifics (McCarthy-Neumann and Kobe 2010a; McCarthy-Neumann and Ibáñez 2012).

The plasticity of infection suggests that seedlings establishing near a heterospecific adult tree may be susceptible to the soil-pathogens accumulated there, and that the overall effects may vary from species to species of host tree and among species of recruiting seedlings. Still, few studies have investigated how different species of seedlings respond to soil-pathogens accumulated underneath heterospecific adult trees (McCarthy-Neumann and Kobe 2010b; Hersh 2009; Mangan et al. 2010; McCarthy-

Neumann and Ibáñez 2012). While some studies have focused on the source of PSF on seedling mortality of particular species, they did not measure the variable response between conspecific vs. heterospecific seedlings in the same soil. This step is critical for assessing whether PSFs are actually operating to produce JC effects and ultimately maintain species coexistence.

Though several studies have linked greenhouse PSF experiments to field experiments (McCarthy-Neumann and Ibáñez 2012), few field studies have seen the same strong responses of PSFs in the field (Klironomos 2002; Kulmatiski et al. 2008). *In situ* studies may provide more ecologically meaningful settings for assessing the effects of PSFs during seedling recruitment (Gustafson and Casper 2004; Hood, Swaine, and Mason 2004; Reinhart et al. 2005; Casper and Castelli 2007).

In this study we investigated the effects of multiple host species' soil on the seedling survival of four species to compare their range of survivorship between conspecific and heterospecific soils. We conducted an *in situ* planting experiment to measure the effects that PSFs may have on seedling survival of several ecologically important tree species. We were able to elucidate the effects of species-specific pathogens affecting both conspecific and heterospecific seedling survival by accounting for varying biotic and abiotic factors such as soil moisture, light levels and the effect of the soil community associated to each adult tree species.

Specific questions we pursued were: 1) does the soil-biotic community associated with specific adult tree species influence recruitment dynamics in these forests? 2) Are soil pathogens shaping recruitment patterns? 3) What are the relative differences and

strengths of the effects? Understanding the complexities associated with the recruitment of dominant tree species in temperate forests is critical in order to predict forest dynamics and community structure.

METHODS

Field Site

Two forest sites were chosen to compare seedling survivorship under the canopy of different host species. Site one was located in Radrick forest, a 14 hectare forest reserve composed of upland mixed Oak-Hickory forest community in Washtenaw County, Michigan (42° 19' N, -83° 24' W) (Hammit & Barnes 1989). The second site was located in the E.S. George Reserve (ESGR) in Livingston County, Michigan (42° 28' N, -84° 00' W). The ESGR is a 525-acre property composed of wetlands, old-fields, and mixed hardwood forests (Hammit and Barnes 1989; Warner 2012). Both sites have sandy loam Alfisols which experience similar precipitation patterns (~762mm per year). Average minimum and maximum temperatures for both sites were -7.7 in January and 36.2 ºC in July (Enloe 2013). Soil nutrient data collected in 2009 found that Nitrogen, Phosphorus, Magnesium, Potassium as well as pH were similar between both sites; see McCarthy-Neumann & Ibáñez (2012).

Seedling species selection & germination

We used four species of seedlings for this study: *Quercus rubra, Acer saccharum, Prunus serotina,* and *Elaeagnus umbellata* (Table 1). All of these species are commonly found within our study sites. *Quercus rubra, A. saccharum,* and *P. serotina* are dominant

species at both locations. *Prunus serotina* has also been a particular focus species of previous PSF studies in which *P. serotina* seedlings exhibited strong negative responses to soil biota extracted from conspecific adult trees (Packer and Clay 2000; Reinhart et al. 2005; Reinhart and Clay 2009). *Elaeagnus umbellata* is an invasive shrub species which competes with the other three tree species (Orr et al., 2005). We purchased *A. saccharum* and *E. umbellata* seeds extracted from wild populations from New Forest Services Co. (Manistee, Michigan, USA), and *Q. rubra* and *P. serotina* seeds, also from wild populations, were purchased from Wildtype, Ltd. (Mason, Michigan, USA) in the fall of 2011 and 2012. Seeds were surface sanitized (0.6% bleach solution) before both scarification and stratification. Seeds were germinated in potting soil in large containers (Metro-Mix 380, Sun Gro Horticulture Canada; Vancouver, British Columbia, Canada).

Seedling preparation & measurements

After germination, seedlings were grown in a greenhouse (Matthaei Botanical Gardens, Ann Arbor, Michigan) for 20-35 days. Before transplanting, we removed the seedlings from the germination containers and lightly removed most of the potting soil from the roots to allow greater exposure of *in situ* soil. We measured initial stem heights to account for differences in survival due to initial size as seedlings with larger seed mass and taller stems tend to have increased survival during the first season (Baraloto et al., 2005).

Planting design

At each site we randomly chose six adult trees consisting of *Q. rubra, A. saccharum*, and *P. serotina* species (*n* = 3 replicates per adult tree species). For the

7

remainder of this paper we will refer to the adult trees as "canopy trees". Adjacent to

each canopy tree we planted rows of tree seedlings comprising individual plots. Half of

each plot consisted of two sub-plots centered at the base of each tree (Fig. 1). Each sub-

plot was formed by four rows of seedlings extending radially (~3.5-4m) from the base of

the tree with one row per seedling species. Multiple replicates from each seedling species

were planted 25cm apart to avoid intraspecific competition ($N_{seedlings\ per\ row} = 7$-15) (see

Fig. 1 for planting schematic). We planted 600 total seedlings in mid-July 2011 and

1,641total seedlings in early May 2012.

Treatment application & survival monitoring

       Seedlings in experimental sub-plots each received 50 mL of a fungicide mixture,

and seedlings in control subplots received equal amounts of tap-water (water was

considered control as the fungicide mixture was water-based). The fungicide solution we

used was a mixture of Medallion and Subdue Maxx (Syngenta, Basel, Switzerland) which

are often used in greenhouses and golf courses to treat damping off disease (Wick 2013).

Treatments were re-applied every two weeks for 16 weeks in order to maintain the

effectiveness of the fungicide during experiment. We carried out the experiment during

the summers of 2011 and 2012. Throughout two field seasons (July to mid-September in

2011 and May to late-August 2012), we recorded seedling survival every two weeks.

Seedlings showing signs of necrosis or desiccation were assigned as dead at each

observation.

Environmental data

During each seedling census, we measured soil moisture in each plot with a Field

Scout TDR 300 soil moisture meter (Spectrum Technologies, Ltd., Illinois, US). We used

the plot's mean soil moisture estimate for inclusion in our survival analysis. We also

collected six light measurements at each plot, once per season, using hemispheric canopy

photos (Sigma SD14 camera with 380° fish-eye lens. Canopy photos were analyzed using

Hemiview (Delta-T, Cambridge, UK) software to determine global site factor (GSF)

information (% of full sun light reaching the ground). We used the average of our six

measurements to describe the light environment under each canopy and for both years.

Data Analysis

To determine the effect of PSFs on the survival of the planted seedlings, we used

a count process in a Cox survival model using a Bayesian hierarchal framework

(Andersen and Gill 1982). Seedling survival was analyzed independently for each

seedling species for each individual seedling i at each census time (j). Mortality was

coded as alive ($N_{ij} = 0$) until time of death ($N_{ij} = 1$), with likelihood:

$$N_{ij} \sim Poisson\ (\lambda_{ij})$$

The intensity function, λ, is derived from two factors influencing mortality: 1) the

intrinsic rate of mortality, (i.e. hazard (h)), and 2) the extrinsic rate of mortality risk (μ):

$$\lambda_{ij} = h_j\ e^{\mu ij}$$

Hazard for each seedling species simulates the temporal variation of mortality that

is not accounted for in the risk function. Hazard was estimated for each census time, h_j,

from a gamma distribution with non-informative parameter values ($h_j \sim Gamma$ (0.01,

0.5)). Risk, μ_{ij}, was calculated as a function of the covariates:

$\mu_{ij}=\alpha_{species\ canopy\ (i),treatment(i)}+\gamma_1 \bullet standardized\ initial\ stem\ height_i + \gamma_2 \bullet soil$

$moisture_{ij}+\gamma_3 \bullet light_{year(i)}$

The alpha parameters (α_*) estimated the effect of growing under each canopy

species and treatment. The gamma $_{(\gamma_*)}$ parameters account for the effects of other

covariates: light, soil moisture, and initial stem height. All coefficients were estimated

from prior distributions with non-informative parameter values letting the data fully

inform their estimation, α_*, $\gamma_* \sim Normal(0,10000)$.

We evaluated different model combinations of covariates and the inclusion of

random effects for site, year of planting and canopy species. After trying several

combinations, we chose the model with the lowest deviance information criterion (DIC)

and most biological meaning (Spiegelhalter et al. 2002). The final model included soil

moisture and initial stem height as fixed effect covariates and no random effects. We also

estimated predicted survival for each canopy and treatment combination using the

survival function: $S_j = \exp\left(-\int_1^j h\right)^{\exp(\mu_j)}$.

We used OpenBUGS 3.2.1(Lunn et al. 2000), an open-source Bayesian modeling

software, to analyze the data. We ran the model for 60,000 iterations to ensure

convergence of the parameters and then we ran another 10,000 iterations from which

posterior parameter values and predicted survival were estimated. Parameter values were

considered significant when the 97.5% CI around the estimate did not include zero. To

test the differences between adult tree species (canopy) and between control and treatment, we estimated the difference between alpha parameters (e.g., $\alpha_{canopy1, \text{control}}$ - $\alpha_{canopy1, \text{fungicide}}$ or $\alpha_{canopy1, \text{control}}$ - $\alpha_{canopy2, \text{control}}$). Differences between alpha values were considered significant if the 95% CI intervals did not include zero.

The effect of the canopy species' effect of fungicide (EF) upon seedling survival is calculated by comparing the probability density functions of seedling survival under control and fungicide treatment (Garrett and Zeger 2000). Comparisons were made at the average survival under control conditions, and EF was calculated as the ratio of the areas under the right side of the curves. The probability of survival reaches a maximum at the mean survival of seedlings under control conditions, which represents the 50% probability threshold of living or dying for that species.

RESULTS

Environmental data

The light environments and soil moisture levels were relatively similar at the two sites (Table 2). The variability in light values among our plots and years was low and when included in the model, model accuracy decreased. Thus light values were excluded from the final analysis.

Field Survival

Mortality represented by the hazard function, *h*, appeared to peak a few weeks after planting and again towards the end of the growing season (see appendix A). The effects of initial seedling stem height and soil moisture significantly influenced the risk of

mortality among all four seedling species (Fig. 2). Survival predictions for *Q. rubra* and *P. serotina* seedlings were positively affected by taller initial stem heights, while *A. saccharum* and *E. umbellata* seedlings experienced the opposite effect. All seedling species except *A. saccharum* exhibited positive responses to soil moisture (Fig. 2).

Testing the differences between the α parameters that assessed the effect of canopy species and treatment indicated that no statistically significant differences related to the identity of the canopy species or the treatments on survival for any of the species (Table 3; Fig 3). Although not significant, all species except for *A. saccharum* under *Q. rubra* and *A. saccharum* canopies had higher survival when treated with fungicide.

Some differences were detected when we compared predicted survival at the end of the season for each species under each combination of canopy species (for Effect of Fungicide [EF] values of each species see Table 4). The effects of adding fungicide varied among canopy species and seedling species which may indicate trait characteristics individual to each species studied. Conspecific canopies tended to have detrimental effects on *Q. rubra* and *P. serotina*, where the fungicide treatment increased *Q. rubra*'s probability of reaching the 50% survival (i.e. the 50-50% probability of surviving the growing season) by 32%. For *P. serotina* seedlings, the application of fungicide increased survival probabilities by 92% (Table 4; Fig. 4). Under heterospecific canopies, *P. serotina* seedlings expressed slightly lower EF values. Alternatively, *A. saccharum* seedlings showed the opposite response with greater EF values under heterospecific canopies with the greatest value occurring under *P. serotina* canopies (Table 4). All EF values for *Q. rubra* seedlings were >1 among all canopy species and

12

did not vary substantially, yet the greatest EF trends for this seedling species also occurred under *P. serotina* canopies. The lowest EF values among all seedling species occurred for *E. umbellata* (EF=0.31) when planted underneath *P. serotina* canopies. This effect value for *E. umbellata* doubled underneath *Q. rubra* canopies and quadrupled under *A. saccharum* canopies.

DISCUSSION

To better understand the role plant-soil feedbacks (PSFs) play in tree species recruitment, this study compared the impact of the soil cultivated underneath the canopies of three adult tree species upon the first season survival of four seedling species. We found that seedlings species exhibited varying degrees of survival when observed under control conditions (i.e., baseline conditions) but within a species, model parameters were not significantly affected by the identity of the canopy species or by the application of fungicide. However, comparing survival probability distributions between treatments of each conspecific seedling-canopy species pairing effects of adding fungicide (EF), we found substantial changes with the application of fungicide (either increased or decreased survival). These trends suggest that PSFs affect seedling survival via the fungal community associated with conspecific canopy trees, but these effects are highly idiosyncratic and their magnitude and direction depend on the identities of the canopy tree and the seedling.

Overall patterns of survival

In our study we found that *A. saccharum* and *P. serotina* seedling species tended to have similar survival rates and these were lower in comparison with the other two species, *Q.*

13

rubra and *E. umbellata*. Other studies have shown that these species tend to have lower survival overall compared to the other seedlings in these forests (Hett and Loucks 1971; McCarthy-Neumann and Ibáñez 2012). As a large seeded species, *Q. rubra* is likely to have higher survival probabilities than smaller seeded species in the first year, due to greater abundance of resources in their cotyledons and high growth rates (Baraloto et al. 2005). This response may explain why *Q. rubra* exhibits strong positive survival underneath all canopies and treatments. These traits of fast growth and substantial seed resources may lend a competitive advantage which allows greater tolerance to poor environmental conditions during establishment (Gilman and Watson 1993). Like *Q. rubra, E. umbellata* seedlings expressed higher survival trends among all canopy species and the two treatments. This result was interesting because *E. umbellata* is characterized as a fast growing plant sprouting from a substantially smaller seed than *Q. rubra. Elaeagnus umbellata*'s high survival rates across canopy species and treatments as well as other abiotic factors, such as low soil moisture, may facilitate the invasive nature of this species (Levine et al. 2006; González-Muñoz et al., 2011; Suding et al. 2013).

Effects of Conspecific Canopy Tree Species

When planted underneath conspecific canopy, *P. serotina* seedlings exhibited a low survival rate under control conditions. This pattern was expected as *P. serotina* survival followed the trends described in previous PSF studies (Packer and Clay 2000; Reinhart et al. 2005; Reinhart and Clay 2009; Reinhart et al. 2012). When comparing survival rates of *P. serotina* seedlings treated with fungicide, we observed an increase of survival rates. Furthermore, the effect of the fungicide (EF) strongly increased the probability of reaching

14

50% survival almost two fold (Fig. 5). Thus, we can infer that the effects of negative PSFs due to fungal pathogens were weak for this species but still detectable in this study.

When planted underneath a conspecific canopy, *Q. rubra* seedlings EF slightly increased survival probabilities relative to the control treatment indicating potential negative PSFs occur for this species (Fig. 5). *Acer saccharum* seedlings, however, experienced an EF reducing survival by 42%. This response indicates that *A. saccharum* canopies lend positive PSFs for conspecific seedlings, an effect observed in a previous study (McCarthy-Neumann and Ibáñez 2013).

Alternating trends between species illustrate how PSF effects can vary in direction and magnitude in conspecific pairings. *Quercus rubra* seedlings experienced weak negative PSF response underneath its conspecific canopy unlike *A. saccharum* which may have been promoted by the fungal community associated with its conspecific canopy. Canopies species which negatively affect conspecific seedlings may potentially promote diversity by decreasing the success of conspecific seedlings growing into monotypic stands. However, tree species which actively promote the survival of conspecific seedlings may not yield the same diversity patterns. A pattern of conspecific promotion will likely develop patchy distributions of *A. saccharum* compared to *Q. rubra* and *P. serotina* as *A. saccharum* seedlings are likely to experience greater survival near a conspecific tree.

Effects of Heterospecific Canopy tree species

Soils associated with *Q. rubra* and *A. saccharum* canopies expressed a lower trend of harmful effects for all heterospecific seedling species (Table 4). Under heterospecific canopies, *P. serotina* seedlings were the most negatively affected seedling species

15

underneath all canopies. *Acer saccharum* seedlings tended to have lower survival rates when planted away from its conspecific canopy. When associated with *P. serotina* canopies, *A. saccharum* seedling mortality more than doubled the EF ratios found when compared to *A. saccharum* EF ratios under other canopy species. *Quercus rubra* did not show strong differences of survival between heterospecific canopies, yet all canopy species expressed similar EF values >1 indicating a detrimental effect of the fungal community for this seedling species. This uniform response indicates that *Q. rubra* is equally affected by both *A. saccharum* and *P. serotina* canopies as well as its own conspecific canopy. Though we did not test differences of *E. umbellata* seedling responses to its conspecific canopy, we found differing EF values among the three heterospecific canopies we studied with a ranked effect. *Elaeagnus umbellata* expressed the lowest EFC values, a beneficial effect of the fungal community, underneath *P. serotina* and *A. saccharum* canopies. These responses were interesting in that all the native species expressed the opposite trends in EF values under these two canopy species.

Plant-soil feedbacks & soil moisture

Soil moisture, was a significant variable affecting survival for all four seedling species. The summer of 2012 was one of the greatest drought events in the region over the last 25 years (Andresen 2012). Long lasting, above-average temperatures occurred throughout the field season with little respite of rain as 214 mm fell during growing season (48.7% less than 2011) (Enloe 2013). But, *A. saccharum,* a moderately drought tolerant species (Hett and Loucks 1971; Caspersen and Kobe 2001) was negatively affected by high levels of soil moisture. Although we did not observe a significantly different effect of the

16

fungicide treatment, this poor survival might have been due to a greater incidence of soil pathogens, but additional study would be needed to test this conjecture.

Overall, the effect of drought was severe in both sites and this phenomenon may have affected the evidence and magnitude of PSF's altogether. Fungal communities are dependent on soil moisture to develop in great number and density (Bell et al., 2006). Therefore pathogenic and mutualistic fungal communities may not have developed in large enough densities to have had a strong effect due to drought. Other studies have found that differences of survival between treatments were dampened under arid conditions as fungal pathogens tend to thrive under moist conditions (Meijer et al., 2011; Reinhart et al., 2012). Thus the low soil moisture levels experience during the second summer (2012) may have greatly mitigated the effects of PSFs in this study.

Role of PSFs in forest diversity

The effect of plant-soil feedbacks are well-detected in greenhouse studies (Klironomos 2002; McCarthy-Neumann and Kobe 2008; McCarthy-Neumann and Ibáñez 2012). In the field, we observed differing responses between seedling species planted underneath conspecific and heterospecific adult trees but survival patterns were not significantly different in all seedling-canopy and treatment combinations. However, we did find alterations of survival probabilities for *P. serotina* seedlings planted under a conspecific canopy due to the application of fungicide. This differential survival of seedlings indicates that PSF can drive survival patterns as described in the JC model and previous PSF studies.

We did not find significant differences of survival between all seedling species (because CI intervals overlapped) for each seedling-canopy species and treatment

17

combinations. Dry environmental conditions and seed resources may play more important roles when predicting survival than PSFs in the first year as seen this study. Previous studies have found that changes in moisture may facilitate regime changes in soil-microbe communities which alter the net effect of PSFs over time (Bell et al., 2006; Meijer et al., 2011). However, we observed considerable differences (ex: *P. serotina* experienced stronger negative feedbacks when planted under conspecific canopies than when planted under hetero specific canopies) of reaching greater probabilities of survival when seedlings were treated with fungicide. These shifts in survival probabilities indicate that a component of each canopy species' associated fungal community has a differential effect on each seedling species. This effect was found to be beneficial (EF <1) for some species but also detrimental (EF>1) for others, and these disparities likely contribute to species coexistence (Ehrenfeld et al., 2005; Mangan et al. 2010; McCarthy-Neumann and Kobe 2010a).

Conclusions

The ultimate likelihood of seedling survival will depend on a given seedling's response to a suite of complex interactions occurring in the area where it establishes(Kotanen 2007; McCarthy-Neumann and Ibáñez 2012; Brandt et al. 2013). By investigating and measuring the effects of driving variables on multiple tree species, we were able to better understand how coexistence could be facilitated in plant communities. Plant-soil feedbacks associated with fungal communities can vary the magnitude of positive or negative effects on seedling success depending on the canopy species and soil moisture regime in the area. Species with poor survival probabilities under all canopy species are unlikely to develop monotypic stands, this negative feedback may facilitate diversity by self-limiting the density

of its own species. Additionally, canopy species which promote other heterospecific

seedlings over their own conspecific seedlings will further enhance heterospecific seedling

success.

LITERATURE CITED:

Agrios, George N. 1988. *Plant Pathology*. San Diego: Academic Press.
      http://hdl.handle.net/2027/[u]: mdp.39015050447310.
Andresen, Jeff. 2012. "Michigan Drought Update for July 12, 2012 | MSU Extension."
      Michigan State University Extention.
      http://msue.anr.msu.edu/news/michigan_drought_update_for_july_12_2012.
Baraloto, Christopher, Pierre-Michel Forget, and Deborah E. Goldberg. 2005. "Seed Mass,
      Seedling Size and Neotropical Tree Seedling Establishment." *Journal of Ecology* 93
      (6): 1156–1166. doi:10.1111/j.1365-2745.2005.01041.x.
Bartelt-Ryser, Janine, Jasmin Joshi, Bernhard Schmid, Helmut Brandl, and Teri Balser. 2005.
      "Soil Feedbacks of Plant Diversity on Soil Microbial Communities and Subsequent
      Plant Growth." *Perspectives in Plant Ecology, Evolution and Systematics* 7 (1)
      (March 31): 27–49. doi:10.1016/j.ppees.2004.11.002.
Bell, Thomas, Robert P. Freckleton, and Owen T. Lewis. 2006. "Plant Pathogens Drive
      Density-dependent Seedling Mortality in a Tropical Tree." *Ecology Letters* 9 (5):
      569–574. doi:10.1111/j.1461-0248.2006.00905.x.
Bever, James D., Ian A. Dickie, Jose M. Facelli, John Klironomos, Mari Moora, Matthias
      Rillig, William D. Stock, Mark Tibbett, and Martin Zobel. 2010. "Rooting Theories
      of Plant Community Ecology in Microbial Interactions." *Trends in Ecology and
      Evoloution* 25: 468–478.
Brandt, Angela J., Hans de Kroon, Heather L. Reynolds, and Jean H. Burns. 2013. "Soil
      Heterogeneity Generated by Plant–soil Feedbacks Has Implications for Species
      Recruitment and Coexistence." *Journal of Ecology* 101 (2): 277–286.
      doi:10.1111/1365-2745.12042.
Casper, Brenda B., and Jeffrey P. Castelli. 2007. "Evaluating Plant–soil Feedback Together
      with Competition in a Serpentine Grassland." *Ecology Letters* 10 (5): 394–400.
Caspersen, John P., and Richard K. Kobe. 2001. "Interspecific Variation in Sapling Mortality
      in Relation to Growth and Soil Moisture." *Oikos* 92 (1) (January 1): 160–168.
Connell, J.H. 1971. "Dynamics in Population". Center for Agricultural Publishing and
      Documentation.
Ehrenfeld, Joan G., Beth Ravi, and Kenneth Elgersma. 2005. "Feedback in the Plant-Soil
      System." *Annual Review of Environment and Resources* 30: 75–115.
      doi:10.1146/annurev.energy.30.050504.144212.
Ehrenfeld, Joan G., Beth Ravit, and Kenneth Elgersma. 2005. "FEEDBACK IN THE
      PLANT-SOIL SYSTEM." *Annual Review of Environment and Resources* 30

(November 21): 75–115. doi:10.1146/annurev.energy.30.050504.144212.

Enloe, Jesse. 2013. "Michigan, Precipitation, February-May 1895-2012". Federal. *NOAA National Climatic Data Center*. http://www.ncdc.noaa.gov/temp-and-precip/time-series/index.php?parameter=pcp&month=5&year=2012&filter=4&state=20&div=0.

Garrett, Elizabeth S., and Scott L. Zeger. 2000. "Latent Class Model Diagnosis." *Biometrics* 56 (4): 1055–1067. doi:10.1111/j.0006-341X.2000.01055.x.

Gause, Georgii Frantsevich. 1932. "Experimental Studies on the Struggle for Coexistence." *Journal of Experimental Biology* 9: 389–402.

Gilbert, Gregory S., and Campbell O. Webb. 2007. "Phylogenetic Signal in Plant Pathogen-host Range." *Proceedings of the National Academy of Sciences of the United States of America* 104 (12) (March 20): 4979–4983. doi:10.1073/pnas.0607968104.

Gilman, Edward F., and Dennis G. Watson. 1993. "Fact Sheet ST-51 (Acer Saccharum, Prunus Serotina, Quercus Rubra, Elaeagnus Umbellata)". Agricultural Engineering Department, Cooperative Extension Service, Institute of Food and Agricultural Sciences, University of Florida, Gainsville FL 32611.

González-Muñoz, Noelia, Pilar Castro-Díez, and Natalia Fierro-Brunnenmeister. 2011. "Establishment Success of Coexisting Native and Exotic Trees Under an Experimental Gradient of Irradiance and Soil Moisture." *Environmental Management* 48 (4) (October 1): 764–773. doi:10.1007/s00267-011-9731-3.

Gustafson, Danny J., and Brenda B. Casper. 2004. "Nutrient Addition Affects AM Fungal Performance and Expression of Plant/fungal Feedback in Three Serpentine Grasses." *Plant and Soil* 259 (1-2) (February 1): 9–17. doi:10.1023/B:PLSO.0000020936.56786.a4.

Hammit, W.E., and B.V. Barnes. 1989. "Composition and Structure of an Old-growth Oak-Hickory Forest in Southern Michigan Over 20 Years." *7th Central Hardwood Forest Conference, Carbondale, IL.*

Hersh, Michelle Heather. 2009. "The role of seedling pathogens in temperate forest dynamics". Ph.D., United States -- North Carolina: Duke University. http://search.proquest.com/docview/304882522/abstract?accountid=14667.

Hett, Joan M., and Orie L. Loucks. 1971. "Sugar Maple (Acer Saccharum Marsh.) Seedling Mortality." *Journal of Ecology* 59 (2) (July 1): 507–520. doi:10.2307/2258328.

Hood, L. A., M. D. Swaine, and P. A. Mason. 2004. "The Influence of Spatial Patterns of Damping-off Disease and Arbuscular Mycorrhizal Colonization on Tree Seedling Establishment in Ghanaian Tropical Forest Soil." *Journal of Ecology* 92 (5) (October 1): 816–823. doi:10.2307/3599380.

Klironomos, John N. 2002. "Feedback with soil biota contributes to plant rarity and invasiveness in communities." *Nature* 417 (6884) (May 2): 67–70. doi:http://dx.doi.org/10.1038/417067a.

Kotanen, Peter M. 2007. "Effects of Fungal Seed Pathogens Under Conspecific and Heterospecific Trees in a Temperate Forest." *Canadian Journal of Botany* 85 (10) (October): 918–925.

Kulmatiski, Andrew, Karen H. Beard, John R. Stevens, and Stephanie M. Cobbold. 2008. "Plant–soil Feedbacks: a Meta-analytical Review." *Ecology Letters* 11: 980–992.

Levine, Jonathan M, Elizaveta Pachepsky, Bruce E Kendall, Stephanie G Yelenik, and

Janneke Hille Ris Lambers. 2006. "Plant–soil Feedbacks and Invasive Spread." *Ecology Letters* 9 (9) (September 1): 1005–1014. doi:10.1111/j.1461-0248.2006.00949.x.

Liu, Yu, Shixiao Yu, Zhi-Ping Xie, and Christian Staehelin. 2012. "Analysis of a Negative Plant–soil Feedback in a Subtropical Monsoon Forest." *Journal of Ecology* 100 (4): 1019–1028. doi:10.1111/j.1365-2745.2012.01953.x.

Lunn, David J., Andrew Thomas, Nicky Best, and David Spiegelhalter. 2000. "WinBUGS - A Bayesian Modelling Framework: Concepts, Structure, and Extensibility." *Statistics and Computing* 10 (4): 325–337. doi:10.1023/A:1008929526011.

Mangan, Scott A., Stefan A. Schnitzer, Edward A. Herre, Keenan M.L. Mack, Mariana C. Valencia, Evelyn I. Sanchez, and James D. Bever. 2010. "Negative Plant-soil Feedback Predicts Tree-species Relative Abundance in a Tropical Forest." *Nature* 466 (August 5): 752–756. doi:10.1038/nature09273.

McCarthy-Neumann, Sarah, and Inés Ibáñez. 2012. "Tree Range Expansion May Be Enhanced by Escape from Negative Plant–soil Feedbacks." *Ecology* 93 (12) (July 17): 2637–2649. doi:10.1890/11-2281.1.

McCarthy-Neumann, Sarah, and Inés Ibáñez . 2013. "Plant-soil Feedback Link Negative Distance Dependence and Light Gradient Partitioning During Tree Seedling Establishment." *Ecology* (January 8). doi:10.1890/12-1338.1. http://www.esajournals.org/doi/abs/10.1890/12-1338.1.

McCarthy-Neumann, Sarah, and Richard K. Kobe. 2008. "Tolerance of Soil Pathogens Co-varies with Shade Tolerance Across Species of Tropical Tree Seedlings." *Ecology* 89 (7): 1883–1892.

McCarthy-Neumann, Sarah, and Richard K. Kobe. 2010a. "Conspecific Plant–soil Feedbacks Reduce Survivorship and Growth of Tropical Tree Seedlings." *Journal of Ecology* 98 (2): 396–407. doi:10.1111/j.1365-2745.2009.01619.x.

McCarthy-Neumann, Sarah, and Richard K. Kobe. 2010b. "Conspecific and Heterospecific Plant–soil Feedbacks Influence Survivorship and Growth of Temperate Tree Seedlings." *Journal of Ecology* 98 (2): 408–418. doi:10.1111/j.1365-2745.2009.01620.x.

Meijer, Seline S., Milena Holmgren, and Wim H. Van der Putten. 2011. "Effects of Plant–soil Feedback on Tree Seedling Growth Under Arid Conditions." *Journal of Plant Ecology* 4 (4) (December 1): 193–200. doi:10.1093/jpe/rtr011.

Mills, Katherine E., and James D. Bever. 1998. "Maintenance of Diversity Within Plant Communities: Soil Pathogens as Agents of Negative Feedback." *Ecology* 79 (5) (July 1): 1595–1601. doi:10.2307/176779.

Orr, Samuel P., Jennifer A. Rudgers, and Keith Clay. 2005. "Invasive Plants Can Inhibit Native Tree Seedlings: Testing Potential Allelopathic Mechanisms." *Plant Ecology* 181 (2) (January 1): 153–165. doi:10.2307/20146831.

Packer, Alissa, and Keith Clay. 2000. "Soil Pathogens and Spatial Patterns of Seedling Mortality in a Temperate Tree." *Nature* 404: 278–281.

Porter, Warren P., and David M. Gates. 1969. "Thermodynamic Equilibria of Animals with Environment." *Ecological Monographs* 39 (3) (July 1): 227–244.

Van der Putten, W. H., C. Van Dijk, and B. a. M. Peters. 1993. "Plant-specific Soil-borne

Diseases Contribute to Succession in Foredune Vegetation” 362 (6415) (March 4): 53–56. doi:10.1038/362053a0.

Reinhart, Kurt O., and Keith Clay. 2009. “Spatial Variation in Soil-Borne Disease Dynamics of a Temperate Tree, Prunus Serotina.” *Ecology* 90 (11) (November 1): 2984–2993. doi:10.2307/25592840.

Reinhart, Kurt O., Daniel Johnson, and Keith Clay. 2012. “Conspecific Plant-Soil Feedbacks of Temperate Tree Species in the Southern Appalachians, USA.” *PLoS ONE* 7 (7) (July 10): e40680. doi:10.1371/journal.pone.0040680.

Reinhart, Kurt O., Alejandro A. Royo, Wim H. van der Putten, and Keith Clay. 2005. “Soil Feedback and Pathogen Activity in Prunus Serotina Throughout Its Native Range.” *Journal of Ecology* 93 (5) (October 1): 890–898. doi:10.2307/3599516.

Spiegelhalter, David J., Nicola G. Best, Bradley P. Carlin, and Angelika van der Linde. 2002. “Bayesian Measures of Model Complexity and Fit.” *Journal of the Royal Statistical Society. Series B (Statistical Methodology)* 64 (4) (January 1): 583–639. doi:10.2307/3088806.

Suding, Katharine N., William Stanley Harpole, Tadashi Fukami, Andrew Kulmatiski, Andrew S. MacDougall, Claudia Stein, and Wim H. van der Putten. 2013. “Consequences of Plant–soil Feedbacks in Invasion.” *Journal of Ecology* 101 (2): 298–308. doi:10.1111/1365-2745.12057.

Warner, Earl. 2012. “Edwin S. George Reserve: Data Resources.” *Edwin S. George Reserve Data Resources*. http://www.sitemaker.umich.edu/esgr/data_resources.

Wick, Robert L. 2013. “Damping-Off of Bedding Plants and Vegetables”. Fact-sheet. *Extension University of Massachusetts*. http://extension.umass.edu/floriculture/fact-sheets/damping-bedding-plants-and-vegetables.

Young, Raymond Allan, and Ronald J. Giese. 1990. *Introduction to Forest Science*. 2nd ed. New York: Wiley. http://hdl.handle.net/2027/[u]: mdp.39015017742118.

TABLES

Table 1. Study species, years planted, native vs. exotic, habit, shade tolerance, and drought tolerance.

Species name (code)	Summer planted	Native vs. Exotic	Habit	Shade tolerance	Drought tolerance
Quercus rubra (Quru)	2011, 2012	native	tree	intermediate	high
Acer saccharum (Acsa)	2012	native	tree	very tolerant	moderate
Prunus serotina (Prse)	2012	native	tree	intolerant	high
Elaeagnus umbellata (Elum)	2011, 2012	exotic	shrub	intolerant	moderate

Notes: Shade tolerance data are from (Burns and Honkala 1990). Drought tolerance data are from (Gilman and Watson 1993).

Table 2: Abiotic factors (mean± 1SD) varied slightly between sites with no significant differences.

Abiotic Data	Light (GSF)*	Soil Moisture % (VWC)**
Radrick	0.07 ± 0.02	14.04 ± 9.93
ESGR	0.084 ± 0.01	13.50 ± 6.93

* Global site factor

** Volumetric Water Content (%)

Table 3. Alpha parameter values (posterior mean & 97.5% CI) for each seedling-canopy species and treatment combination. Tests indicated that alphas did not vary between canopy species or the treatment among the seedling species.

Seedling species	*Q. rubra* Canopy		*A. saccharum* Canopy		*P. serotina* Canopy	
	Control	Fungicide	Control	Fungicide	Control	Fungicide
Quru	-2.8 (-3.6, 1.9)	-2.8 (-3.6 1.7)	-2.6 (-3.3, -1.7)	-2.6 (-3.4, -1.7)	-2.6 (-3.4, -1.8)	-2.7 (-3.5, -1.8)
Acsa	-2.6 (-3.5, -1.7)	-2.5 (-3.4, -1.7)	-2.7 (-3.6, -1.8)	-2.7 (-3.6, -1.8)	-2.7 (-3.5, -1.7)	-2.8 (-3.7, -1.9)
Prse	-2.0 (-2.8, -1.2)	-2.1 (-2.9, -1.2)	-1.8 (-2.5, -0.9)	-1.8 (-2.6, -1.0)	-1.7 (-2.6, -0.8)	-2.0 (-2.9, -1.1)
Elum	-2.5 (-3.4, -1.6)	-2.4 (-3.3, -1.5)	-2.5 (-3.4, -1.6)	-2.6 (-3.5, -1.7)	-2.6 (-3.5, -1.7)	-2.4 (-3.3, -1.5)

Table 4. Effects of conspecific canopy fungal community (EFC) upon seedling survival under non-fungicide conditions vary between species. Values >1 indicate a detrimental effect on seedling survival, and values <1 indicate a beneficial effect.

Seedling species	*Q. Rubra* Canopy	*A. saccharum* Canopy	*P. serotina* Canopy
Quru	1.32	1.2	1.38
Acsa	0.6	0.58	1.45
Prse	1.38	1.27	1.92
Elum	0.65	1.41	0.31
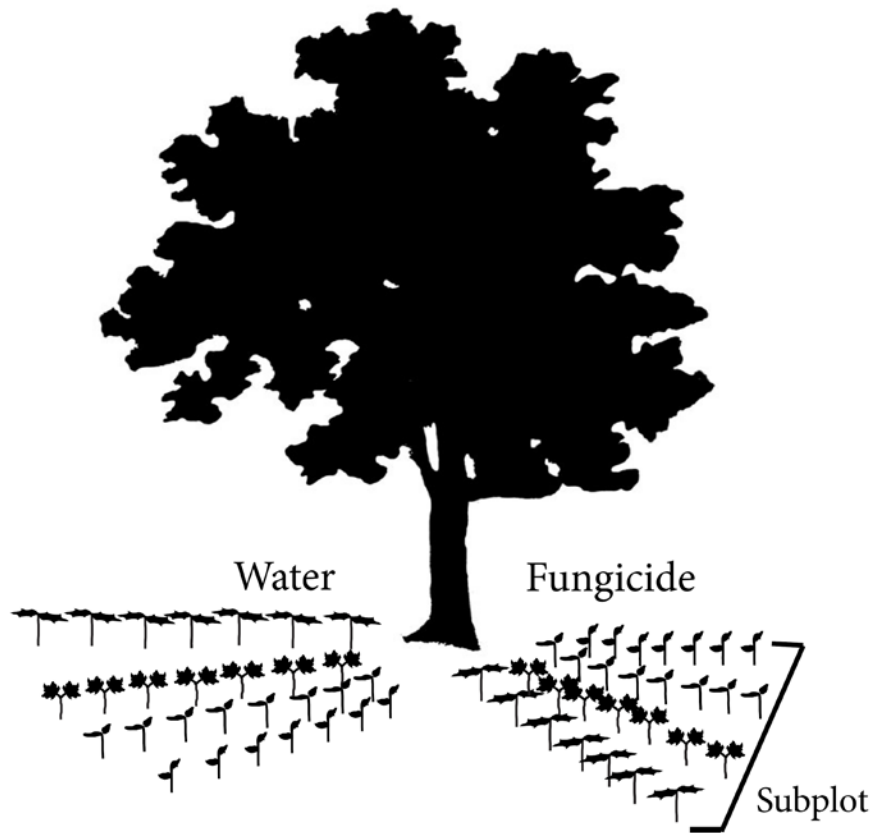
Figure 1. Planting design of each plot with two sub-plots of seedlings extending radially from the base of the adult tree. One treatment was applied per subplot every two weeks during the growing season. (Adult tree image: Hatzigeorgiou 2013).
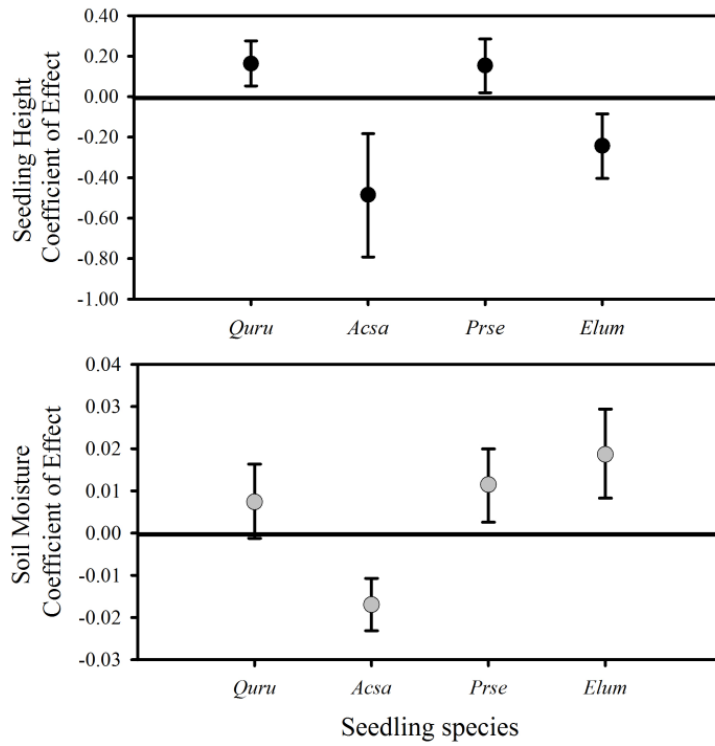
Figure 2. Effects of initial seedling height and soil moisture coefficients (mean ± 95% CI) significantly influenced seedling survival. *Quercus rubra* and *E. umbellata* commonly express positive responses to both initial stem height and soil moisture. Parameter values have been multiplied by -1 to represent their effect on survival as the model estimates mortality.
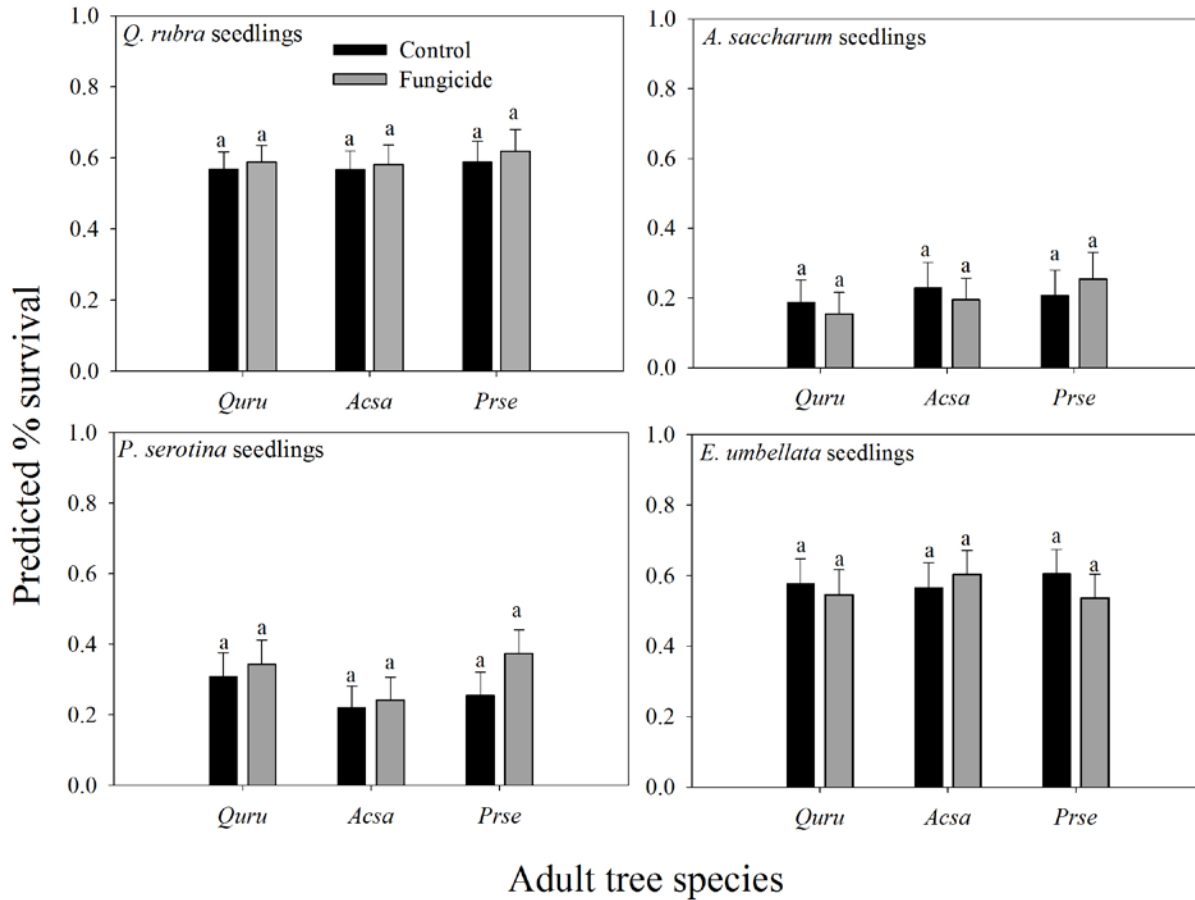
Figure 3. Predicted seedling species survival (mean ± 95% CI) did not alter among the three canopy tree species. Additionally, no differences were detected between fungicide and control treatments. Letters indicate significant differences. Quru: *Q. rubra*, AcSa: *A. saccharum*, Prse: *P. serotina*.
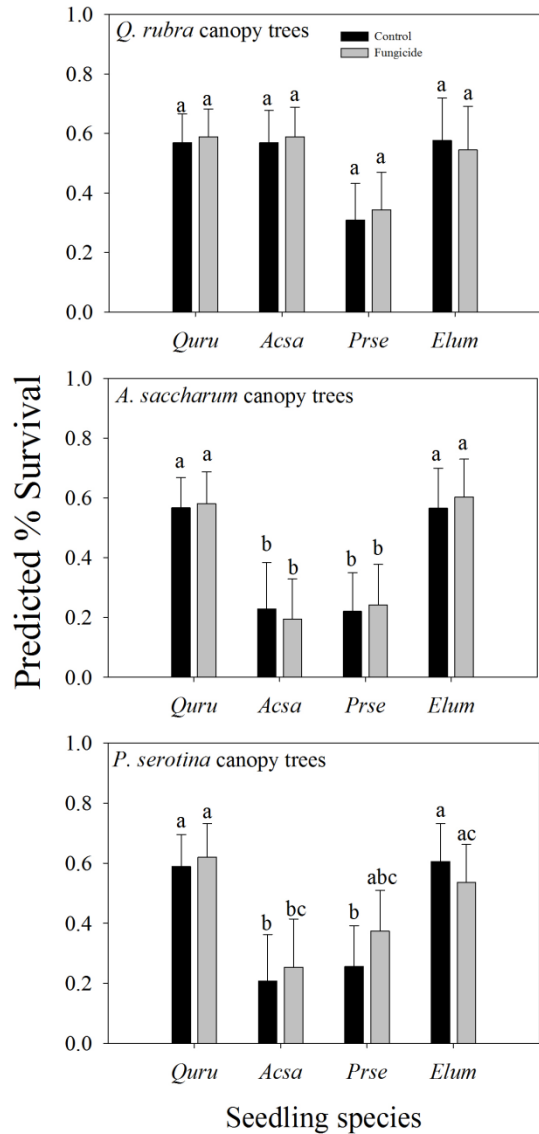
Figure 4. Ranking of seedling survival (mean & 95% CI) varied slightly depending on canopy tree species. *Quercus rubra* and *E. umbellata* appeared to be favored underneath all canopy species but survival was not uniformly significant. Letters indicate significant differences between seedling species and treatments.
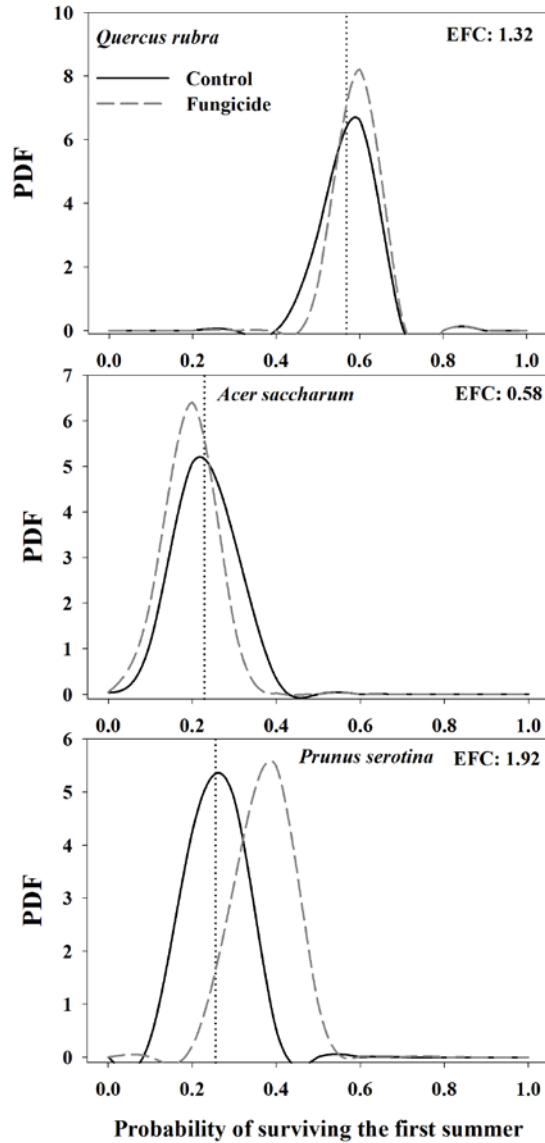
Figure 5. Probability density functions (PDF) of seedling survival when growing under conspecific canopies, under control conditions (solid line) and under fungicide treatment (dashed lines). Vertical line indicates the average survival under control conditions. The effect of the fungal community (EF) represents the increase (>1) or decrease (<1) caused by the fungicide treatment on the probability of surviving to the control average or higher.

APPENDIX

A. Seedling hazard curves for all seedling and canopy tree species combinations at both sites across the 2012 field season. Large spikes in the curves indicate when increased hazard occurred during the season.
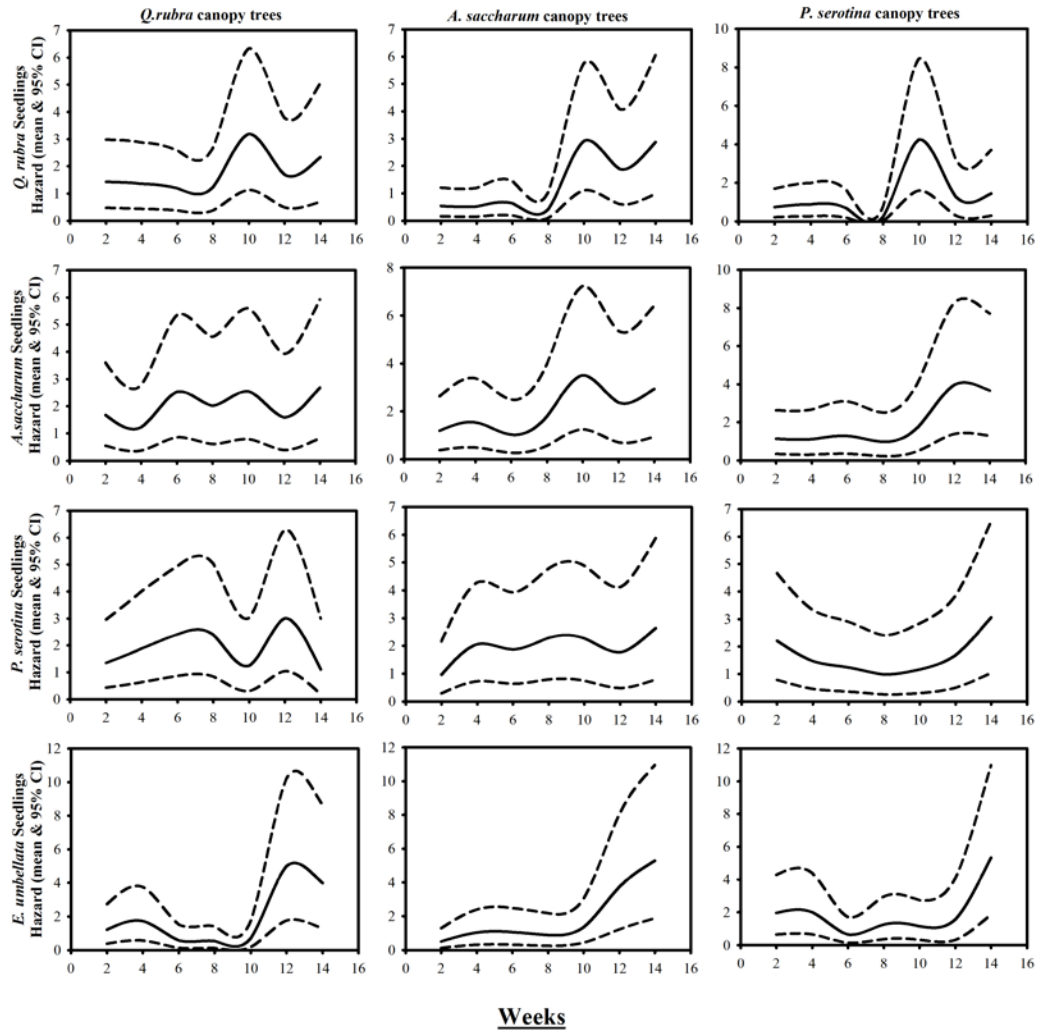
B. Model outputs for gamma parameters representing the coefficient of effect for soil moisture and initial stem height on seedling mortality (mean & 97.5% CI). Coefficients were transformed to reflect effects on survival by multiplying all values by -1.

Seedling Species	Soil Moisture (γ 1)	Initial stem height (γ 2)
Quru	-0.01 (-0.02, 0.001)	-0.2 (-0.3, -0.05)
Acsa	0.02 (0.01,0.02)	0.5 (0.2, 0.8)
Prse	-0.01 (-0.02,-0.003)	-0.2 (-0.3, -0.02)
Elum	-0.02 (-0.03, -0.008)	0.2 (0.09, 0.4)