

Final Report: Improving the Success of Plant Restorations through Analysis of Plant-Microbe Feedback

Brooke Pickett and Emma Aronson
July 2015–June 2016
Department of Plant Pathology
University of California Riverside, CA 92521

Background

It has become clear over time that some of the most fundamental regulators of plant composition are nutrients and microorganisms in the soil (Bever et al., 2010). The fact that plants must take up nutrients and water to survive points to the importance of root symbionts, such as arbuscular mycorrhizal fungi (AMF) and nitrifying bacteria. About 20,000 plant species are dependent on microbes for survival (van der Heijden, Bardgett, & van Straalen, 2008), which means microbial composition of the soil can actually control the plant composition.

Working from this finding, ecologists discovered that introduced plant species have a profound effect on native plant communities. Many sources describe how plant invasion can change abiotic and biotic soil conditions, affecting the soil environment in ways that inhibit the growth of native plants long after the removal of invasive species (Bever et al., 2010; Cuddington, 2012; Eviner, Hoskinson, Hawkes, & Eviner, 2010; Hawkes, Belnap, D'Antonio, & Firestone, 2006; Jordan, Larson, & Huerd, 2007; Kourtev, Ehrenfeld, & Häggblom, 2002). Invasion can alter important nutrient cycles, prevent microbial mutualisms related to water and mineral absorption, change soil food webs, and introduce plant pathogens that inhibit natives (Belnap, Phillips, Sherrod, & Moldenke, 2005; Jordan et al., 2007). The projects I have worked on during the past year have been focused on the microbial composition changes brought on by invasive grasses as well as the ways in which these changes affect the growth of native and invasive plants. This research will help efforts to restore native California sage scrub (CSS), especially in areas where invasive grasses are dominant.

Research Projects

Brachypodium Study

The project I took up this year focuses on understanding how microbe-plant interactions influence the proliferation of invasive grass species. The study is specifically focused on three related species within the genus *Brachypodium*: *B. distachyon*, *B. hybridum*, and *B. stacei*. *Brachypodium distachyon* split into these three species, each with a different chromosome number. It is assumed that *B. hybridum* is widely invasive in California, while *B. distachyon* is not. This study aims to understand the differences in rhizosphere microbial composition between *B. hybridum* and *B.*

distachyon and determine if they change the native soil microbial composition in different ways. This information may help us discover if microbe-plant interactions are promoting the spread of the invasive species.

We collected samples in Spain (*B. hybridum* native range) and California. The California sites sampled this year include: Rancho Jamul, Irvine Ranch, Rancho Marino, Sedgwick, Alganorte Park, Alicante Park, and the Channel Islands. At each sampling site, five *Brachypodium* shoots, five associated roots, five non-*Brachypodium* invasive grass shoots and roots, five bulk soil samples per plant, and five 5-cm soil cores were collected. Soil moisture and temperature were also collected at each soil core site. I am now using high-throughput sequencing (Illumina MiSeq) of PCR-amplified 16S sequences to identify changes in bacterial and archaeal communities over time in the rhizosphere and bulk soils.

Inoculation Experiment

Southern California has a long history of ranching and agriculture that have transformed much of the landscape from chaparral and CSS to nonnative grassland. In Rancho Sierra Vista, Harding grass (*Phalaris aquatica*) is an invasive bunchgrass that was introduced and rapidly expanded to about 25 acres. Removal of the grass began in 2006 and as of 2013 the infestation was down to normal maintenance levels with eradication expected in the next 2 years. However, following removal of the *P. aquatica*, native plants have largely failed to recolonize the areas from which the grass was removed, despite bordering intact CSS. This suggests that the soil conditions have become unfavorable for natives to grow, possibly due to legacy effects of *P. aquatica*.

After a greenhouse study, I discovered that the native plants had different germination rates and biomass when grown in either native soil or post-invasive soil, suggesting the presence of *P. aquatica* legacy effects. I then wanted to know if these differences in native plant growth could be explained by changes in the microbial community, not just a change in the chemical content of the two soils. If so, remediation of soil microbial conditions through inoculation with native soil could improve the restoration success of native plants in areas where invasive grasses previously grew.

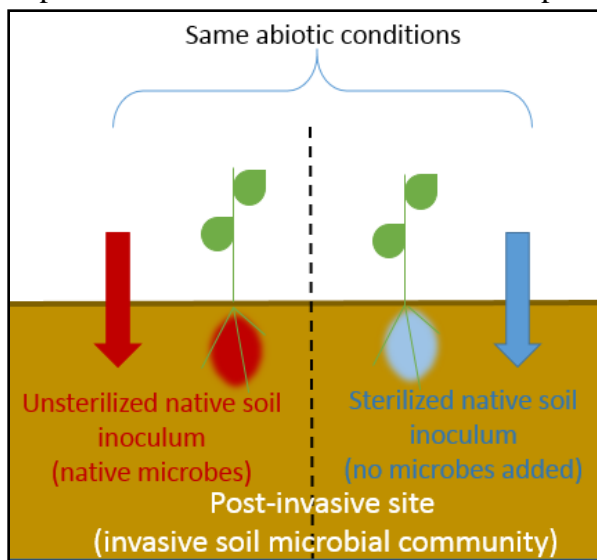


Figure 1. Diagram of field experiment

To answer this question, I grew 3 common native species (*A. californica*, *S. leucophylla*, and *B. pilularis*) in either unsterilized or sterilized native soil before planting them into the 25-acre site (Figure 1). After 7 months of growth, I discovered that native plant growth was significantly increased after inoculation with the unsterilized soil. During the last year this project has been focused on understanding the underlying changes in microbial composition that may be driving these trends in native plant growth. I have used high-throughput sequencing (Illumina MiSeq) to identify the changes in bacterial and archaeal communities over time in

the soil core samples I collected during this experiment.

Restoration Study

My goal with this project was determining which microbes actually associate with native plant roots in a restored system with soil inoculum versus an intact native system (i.e., one that has not been invaded). Further information about the microorganisms that associate with these native plant species in restored versus native sites may better inform restoration methods, increase understanding of the role of microbes in restoration, and ultimately increase restoration success.

I collected native and post-invasive soil from the Motte Rimrock Reserve and harvested seed of the three native plant species. Approximately 3 months prior to the field study, all of the plants ($n = 120$) were grown in a UCR greenhouse in containers filled with sterilized standard potting mix combined with native soil. Half of the plants ($n = 60$) were meant to be transplanted from the containers into an intact stand of California sage scrub (native site) and the other 60 plants were to be transplanted into a site where the grass has been cleared (post-invasive site). A total of 60 plants were to be destructively sampled at the end of this experiment and the other 60 were to remain intact as part of a small restoration. Unfortunately, this experiment has seen several setbacks; after many attempts, few seeds germinated properly, and those that did were too weak to survive. This may be due to the considerable heat wave in Riverside or the age of the seed stock that was used. I will repeat this project in the later part of 2016.

Conclusions

The *Brachypodium* project will improve understanding of how microbe-plant interactions influence the spread of invasive grass species; the inoculation project will demonstrate if and how native plant restoration is affected by post-invasive soil microbial composition; and the restoration study may allow us to determine which microbes associate with native plants in native versus restored systems. The overarching goal of all these studies is to increase native plant survival in restorations by better understanding the role of microbes post-invasion on native plant restoration. One practical application of the findings to date is in the use of native soil inocula in restorations. Instead of just seeding a plot with native seed stock, growing the plants in native soil before transplanting will allow them to form valuable associations with native microbes, leading to improved restoration outcomes.

Works Cited

- Belnap, J., Phillips, S., Sherrod, S., & Moldenke, A. (2005). Soil biota can change after exotic plant invasion: does this affect ecosystem processes? *Ecology*, *86*(11), 3007–3017.
- Bever, J. D., Dickie, I. A., Facelli, E., Facelli, J. M., Klironomos, J., Moora, M., ... Zobel, M. (2010). Rooting theories of plant community ecology in microbial interactions. *Trends in Ecology & Evolution*, *25*(8), 468–478. doi:10.1016/j.tree.2010.05.004
- Cuddington, K. (2012). Legacy Effects: The Persistent Impact of Ecological Interactions. *Biological Theory*, *6*(3), 203–210. doi:10.1007/s13752-012-0027-5

- Eviner, V. T., Hoskinson, S. A., Hawkes, C. V., & Eviner, B. V. T. (2010). Ecosystem impacts of exotic plants can feed back to increase invasion in western US rangelands. *Rangelands*, 32(1), 21–31.
- Hawkes, C. V., Belnap, J., D'Antonio, C., & Firestone, M. K. (2006). Arbuscular Mycorrhizal Assemblages in Native Plant Roots Change in the Presence of Invasive Exotic Grasses. *Plant and Soil*, 281(1-2), 369–380. doi:10.1007/s11104-005-4826-3
- Jordan, N. R., Larson, D. L., & Huerd, S. C. (2007). Soil modification by invasive plants: effects on native and invasive species of mixed-grass prairies. *Biological Invasions*, 10(2), 177–190. doi:10.1007/s10530-007-9121-1
- Kourtev, P. S., Ehrenfeld, J. G., & Häggblom, M. (2002). Exotic Plant Species Alter the Microbial Community Structure and Function in the Soil. *Ecological Society of America*, 83(11), 3152–3166.
- Van der Heijden, M. G. a, Bardgett, R. D., & van Straalen, N. M. (2008). The unseen majority: soil microbes as drivers of plant diversity and productivity in terrestrial ecosystems. *Ecology Letters*, 11(3), 296–310. doi:10.1111/j.1461-0248.2007.01139.x