How Fungicide Alters the Hidden Mycobiome of a Restored Prairie System

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PRECS Phenotypic Plasticity Research Experience for Community College Students

Introduction

- Fungal Endophytes are microscopic fungi that live inside plant tissues and form a symbiotic relationship that influences the fitness of both parties.
- Fungicides are a widely used method of crop disease control in agriculture, but fungicides can be carried into other environments by water and wind.
- This experiment looks at how long-term fungicide exposure affects diversity of fungal endophytes that are grown *in vitro* as well as screens them for phosphate solubilization ability.
- Phosphate is a vital macronutrient that is essential for making nucleic acids (DNA, RNA) as well as playing a vital role in energy transfer throughout the plant's cells. Phosphate solubility allows the plants to develop higher efficiency for water and nutrients use. Microbes that can solubilize phosphate help plants receive readily available phosphate.

Background

- Leaf samples utilized for this experiment were collected from the Philips Tract, a USDA funded field experiment that focuses on how residual agricultural activity impact the restored prairie that has been established on former farmlands. The far-reaching goal for the larger project is to understand how fungicide drift and residual fertilizers affect the symbiotic relationship between fungal endophytes and their host plant.

Center photo taken by Noah Brown of research team collecting leaf samples. Left and right images were taken by Mya Reyes while aiding in Bee tent experiment at Phillips Tract.

Acknowledgments

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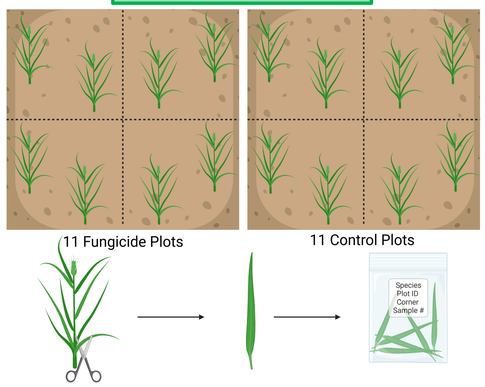
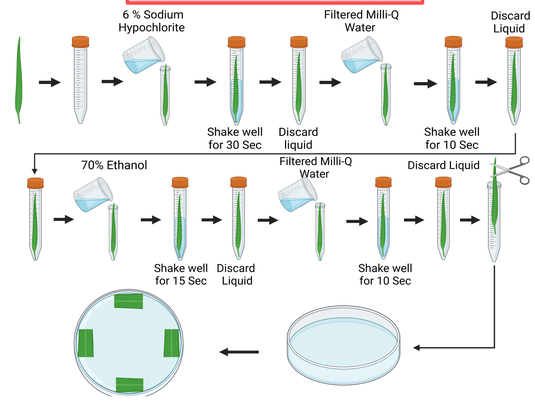
Methods

Leaf Sample Collection

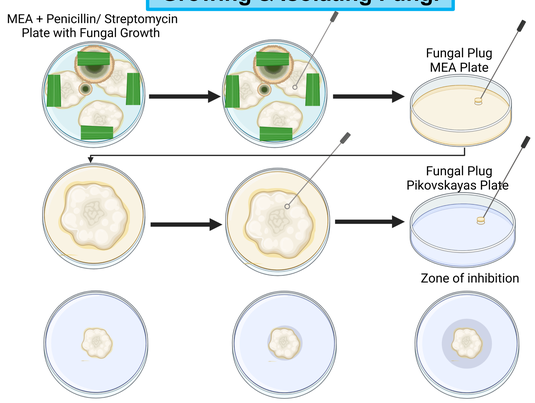


Leaf samples were collected from 2 different plants of the same species per corner of the plot.
Species:
- ***Monarda fistulosa***
- ***Penstemon digitalis***
- ***Pycanthemum virginianum***
- ***Andropogon gerardii***
- ***Sorghastrum nutans***

Surface Sterilizing Leaves



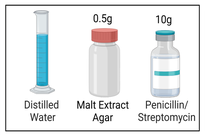
Growing & Isolating Fungi



Fungal Media Procedure

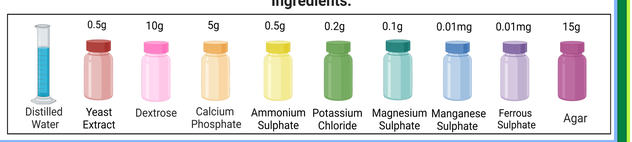
MEA Procedure

Ingredients:



Pikovskayas Procedure

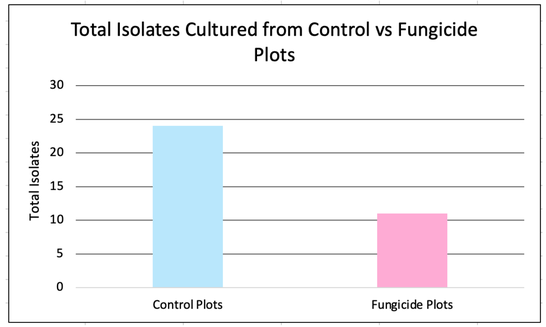
Ingredients:



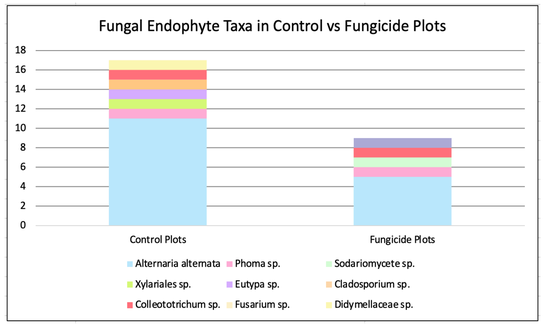
Results

t-Test: Two-Sample Assuming Unequal Variances		
Morphotypes per Leaf		
	Control Plots	*Fungicide Plots*
Mean	**0.571428571**	**0.25**
Variance	0.68989547	0.238372093
Observations	42	44
Hypothesized Mean Diff	0	
df	66	
t Stat	2.174813751	
P(T<=t) one-tail	0.016616387	
t Critical one-tail	1.668270514	
P(T<=t) two-tail	**0.033232774**	
t Critical two-tail	1.996564419	

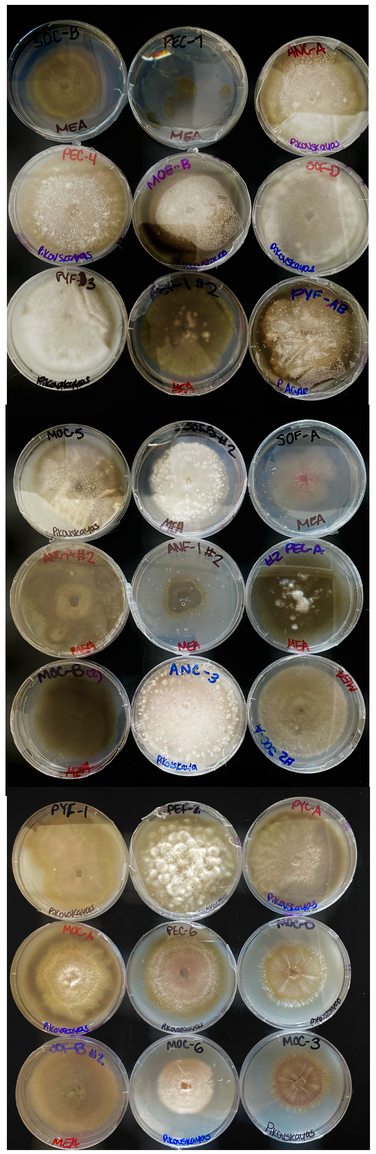
This represents a significant reduction of fungal endophytes on average from fungicide plots.



This graph is a visual of a total difference of endophyte cultures between plots.



This graph shows the difference endophyte compositions from a few sequenced isolates.



This photo is a variety of fungal endophytes grown on Pikovskayas and malt extract agar.

Conclusions

- Long term fungicide exposure in restored prairies significantly reduce their foliar fungal endophyte diversity.
- A lack of phosphate solubilization ability in the initial screening suggest that endophytes may be specialized depending on what plant tissue they inhabit.

Future Work

- Completing DNA sequencing of fungal isolates as well as DNA sequencing from original leaf samples.
- Further metabolite assays to determine how the fungal endophytes influence plant host

References

Rodriguez, R. J., et. al. (2008)
War Farooq Aadil et. al. (2023)
Karlsson Ida et. al. (2014)

I would like to thank the research team (pictured above) for all the help and memories they provided me with.