

# Soil Microbial Communities Under Grapevine Chlorosis

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Dr. Tarah S. Sullivan  
[t.sullivan@wsu.edu](mailto:t.sullivan@wsu.edu)

# About Me...



- Soil Scientist
- Metals Biogeochemistry
- Linking underground processes and organisms to aboveground variables
  - Plant-beneficial microorganisms
- ‘Concord’ work with Joan Davenport in 2014

Microbes → Soil Quality → Soil Health → Plant Health

# Microbes are Critical to Soil Health



# What is “Soil Quality”?

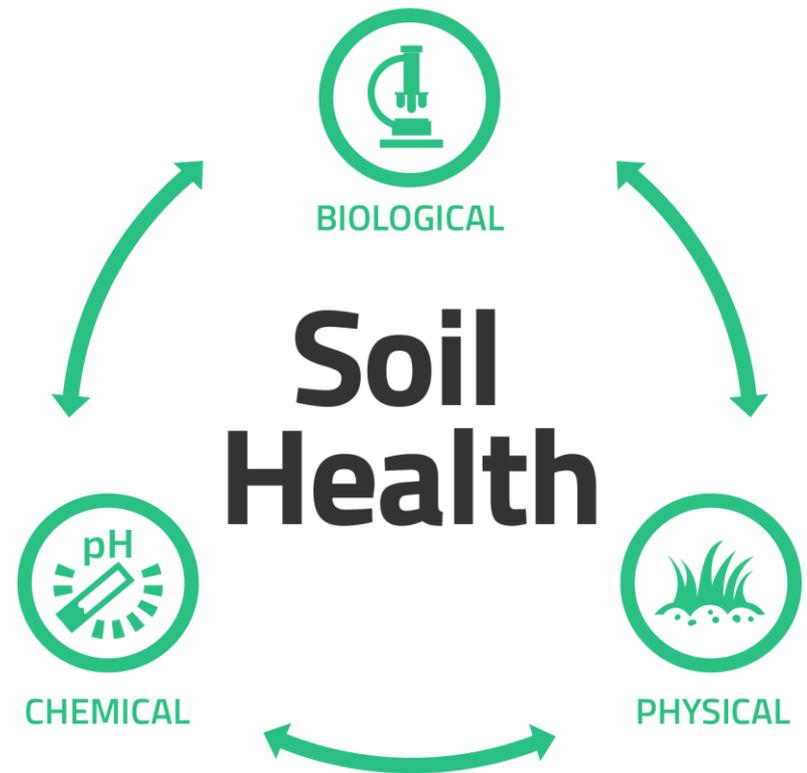
- “The capacity of a specific kind of soil to function within the natural or managed ecosystem boundaries, to sustain plant and animal productivity, maintain or enhance water and air quality, and support human health and habitation.”
- How well soil does what we want it to do...
- Inextricably linked to sustainability – assessing and managing so that soil continues to do what (function) we want it to do.

# What does soil do?

- Regulating water – soil controls where rain, snowmelt, and irrigation water goes.
- Cycling nutrients – C, N, P, macro- and micro-nutrients are stored, transformed, and cycled through soil .
- Filtering potential pollutants – minerals and microbes in soil filter, buffer, degrade, immobilize, and detoxify organic and inorganic materials, including industrial and municipal by-products and atmospheric deposits.
- Sustaining life – the diversity and productivity of living things depends on soil.
  - Grape yield, vine health, berry qualities (“terroir”?)

# Soil Microbes and Soil Health

- Soil **quality** characteristics can often be managed to enhance soil **health**.
  - Physical, chemical and biological factors can be altered to create a hospitable environment for soil life = soil health.



# Biological Soil Health Indicators

- Biomass & community composition.
  - Microbes
  - Earthworms
  - Nematodes
  - Arthropods
- Soil respiration
- Pathogen load
- Microbial nutrient cycling (enzymes & chelators)

## What they can tell us:

- Biodiversity supported.
- Catalytic potential and repository for C and N.
- Soil productivity.
- Microbial activity.
- Rates and pools of nutrient cycling.

# Grapevine Chlorosis

- Symptoms
  - Yellowing of leaves
  - Burning around edges
  - Reduced yield
  - Vine death
- Possible Causes:
  - Nutrient deficiencies (Fe)
  - High soil pH, Ca-P-Fe interactions.
  - Cool, wet springs



# Grapevine Chlorosis

- Solutions:
  - Fe-chelates (Fe-EDDHA)
    - Annual applications



# Question 1



# Q1: Are there differences in the rooting zone community of healthy vs. chlorotic vines?

- 2016 – Sampling May Nursery:
  - Homogenous Silt loam
  - Drip irrigation
  - Paired: healthy vs chlorotic vines
- Surface soils, within ~1ft of vine:
  - Bacterial and fungal sequences
- Leaf tissues at veraison:
  - Nutrient content



# Q1: Leaf tissues and soil

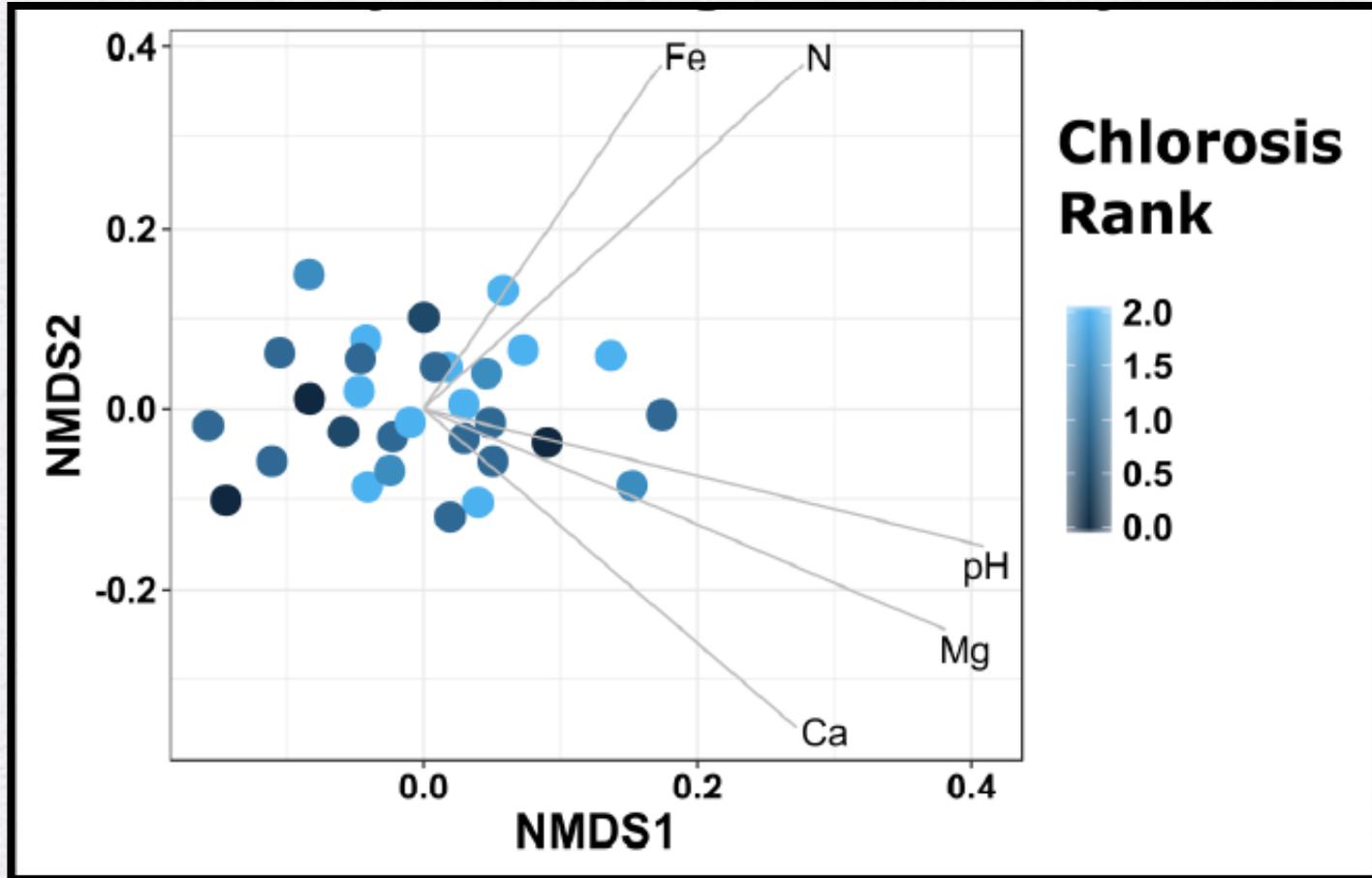
## Correlations Between Measurements:

Can we predict/explain chlorosis severity?

<b>Environmental Factor</b>	<b>Chlorosis Rank</b>	<b>Leaf Mg</b>
<b>Soil Fe</b>	0.01	-0.24
<b>pH</b>	0.23	0.29
<b>Chlorosis Rank</b>	1.00	0.42*
<b>Leaf Mg</b>	0.42*	1.00
<b>Leaf N</b>	0.15	0.14
<b>Leaf P</b>	-0.53*	-0.36
<b>Leaf K</b>	-0.50*	-0.42*
<b>Leaf Ca</b>	0.27	0.64*

Lewis, R., LeTourneau, M.K., Davenport, J. & T.S. Sullivan, 2018. 'Concord' grapevine nutritional status and chlorosis rank associated with fungal and bacterial root zone microbiomes. *Plant Physiology and Biochemistry*, 13 (129): 429-436.

# Q1: Fungal Community



# Q1: Fungal Community

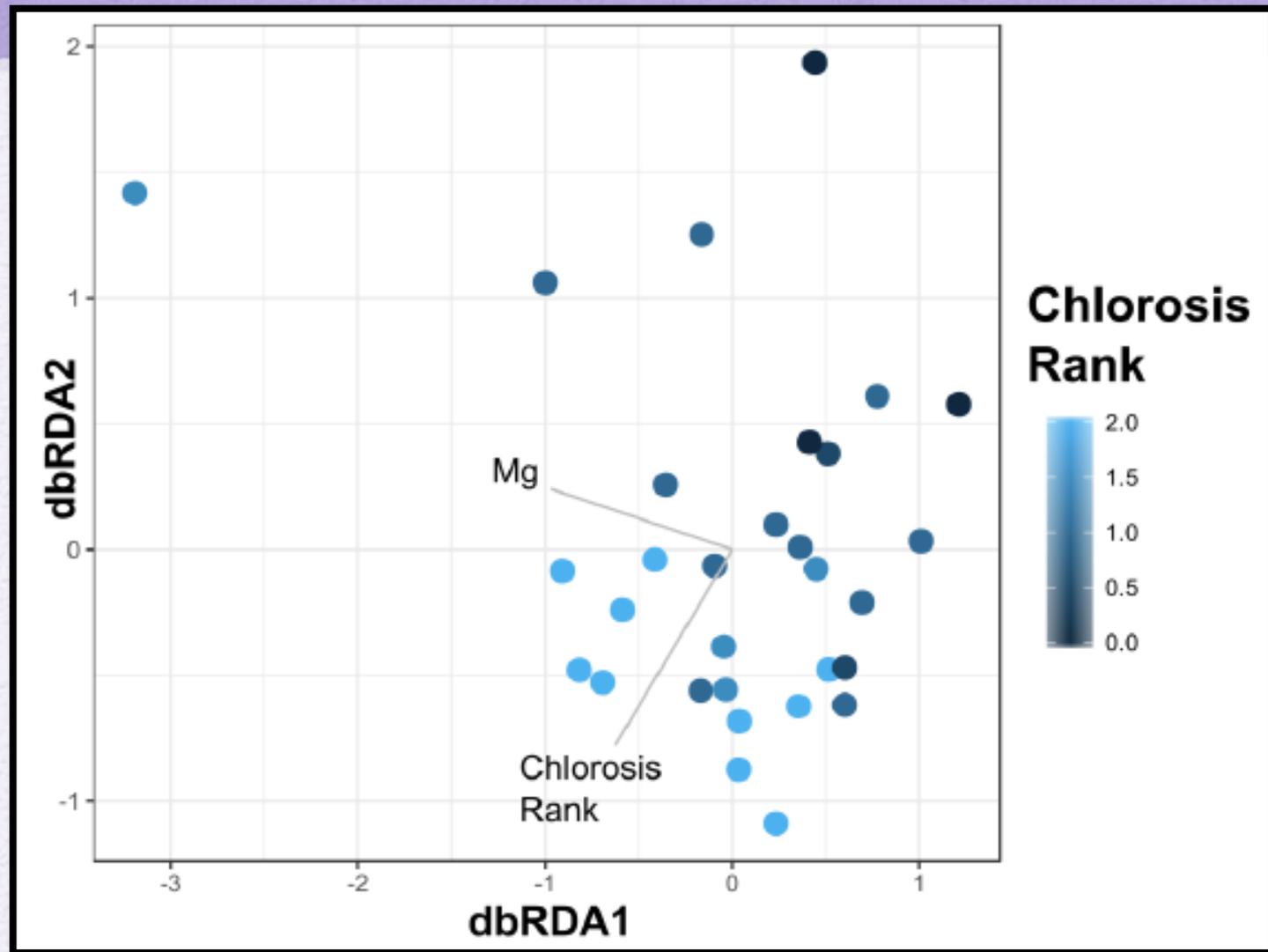
## Leaf Ca

Phylum	Class	Order	Family	Genus	PCC	VE
<i>Ascomycota</i>	<i>Sordariomycetes</i>	<i>Sordariales</i>	<i>Lasiosphaeriaceae</i>	<i>Podospora</i>	0.5	0.3
<i>Ascomycota</i>	<i>Leotiomycetes</i>	<i>Helotiales</i>	unidentified	unidentified	0.5	0.3
<i>Ascomycota</i>	NA	NA	NA	NA	0.5	0.3
<i>Basidiomycota</i>	<i>Wallemiomycetes</i>	<i>Geminibasidiales</i>	<i>Geminibasidiaceae</i>	<i>Geminibasidium</i>	0.5	0.3

## Soil Fe (mg/kg)

Phylum	Class	Order	Family	Genus	PCC	VE
<i>Ascomycota</i>	<i>Sordariomycetes</i>	<i>Hypocreales</i>	<i>Nectriaceae</i>	<i>Fusarium</i>	0.7	0.5
<i>Ascomycota</i>	<i>Eurotiomycetes</i>	<i>Eurotiales</i>	<i>Incertae sedis</i>	<i>Thermomyces</i>	0.6	0.3
<i>Ascomycota</i>	<i>Eurotiomycetes</i>	<i>Chaetothyriales</i>	unidentified	unidentified	0.5	0.3
<i>Ascomycota</i>	<i>Eurotiomycetes</i>	<i>Eurotiales</i>	<i>Trichocomaceae</i>	<i>Penicillium</i>	0.5	0.3
<i>Basidiomycota</i>	<i>Agaricomycetes</i>	<i>Auriculariales</i>	unidentified	unidentified	0.5	0.3

# Q1: Bacterial Community



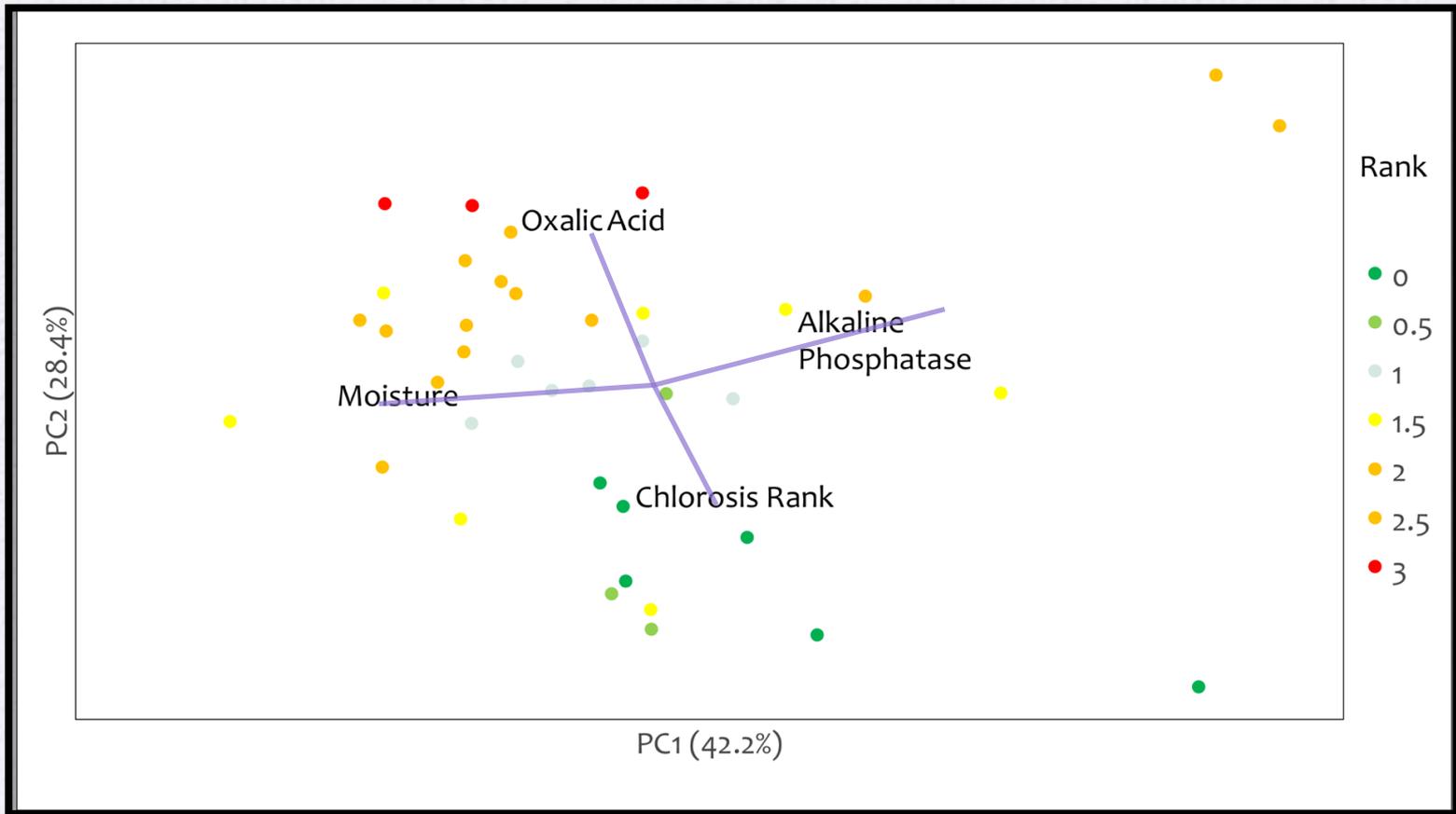
# Q1: Bacterial Community

Families strongly associated with healthy vines:

Phylum	Class	Order	Family	PCC	VE
<i>Proteobacteria</i>	<i>Deltaproteobacteria</i>	<i>Myxococcales</i>	<i>Haliangiaceae</i>	-0.8	0.6
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Rhodospirillales</i>	<i>Rhodospirillaceae</i>	-0.7	0.5
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Actinomycetales</i>	<i>Micromonosporaceae</i>	-0.7	0.5
<i>Bacteroidetes</i>	<i>[Saprospirae]</i>	<i>[Saprospirales]</i>	<i>Chitinophagaceae</i>	-0.7	0.5
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Sphingomonadales</i>	<i>Sphingomonadaceae</i>	0.7	0.5
<i>Bacteroidetes</i>	<i>[Saprospirae]</i>	<i>[Saprospirales]</i>	<i>Chitinophagaceae</i>	-0.7	0.4
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Actinomycetales</i>	<i>Streptomycetaceae</i>	-0.7	0.4

- Only 3 of the top 30 families of bacteria were positively associated with chlorosis.
  - Previously unidentified organisms.
- Grouping due to LACK of beneficial organisms and metabolic capabilities.

# Q1: Bacterial Community Activities



# Q1: Summary

- Are there differences associated with chlorosis?
  - Bacteria: Yes, likely due to altered root exudates
    - Increased oxalic acid production in roots of chlorotic vines.
  - Fungi: Not clear. Soil factors probably a stronger driving factor.
- Notable:
  - No obvious pathogen load on chlorotic vines.
  - C & N metabolism differed between bacteria of chlorotic vs healthy vines.
  - Many plant growth promoting families associated with healthy vines.

# Question 2



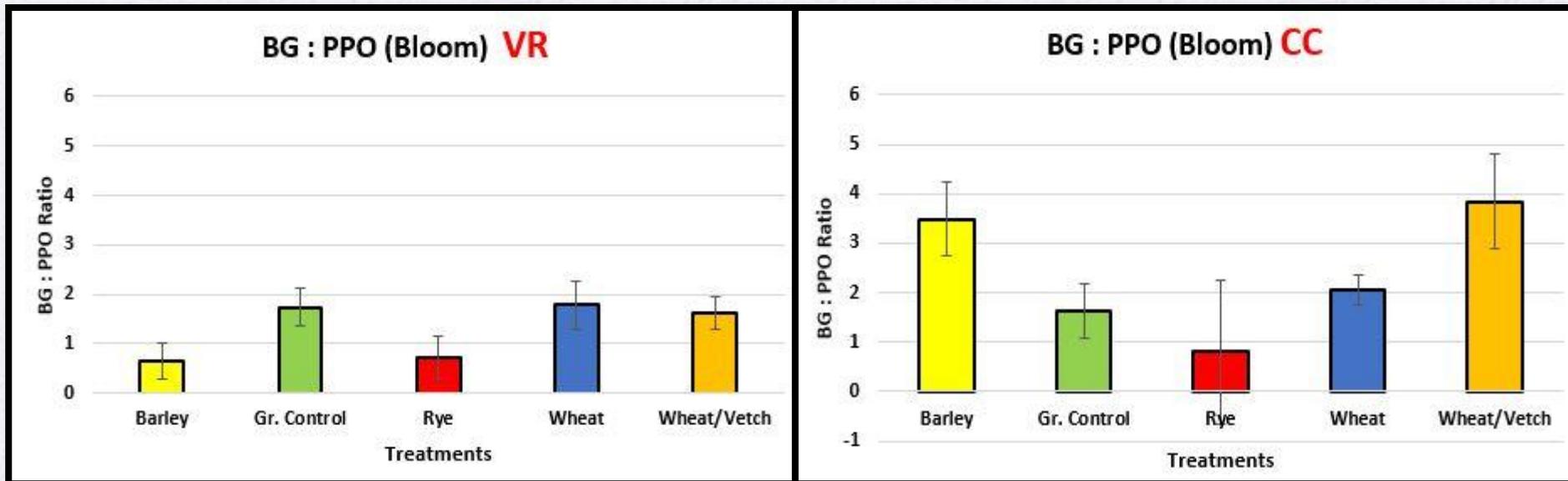
## Q2: Can Inter-Row Cover Crops Increase Microbial Soil Health & Decrease Chlorosis?

- 2017 – Microbial activities, soil health, vine chlorosis.
  - Wheat, Barley, Rye, Wheat/Vetch mix comparison



# Q2: Microbial Activities: Enzyme Activities

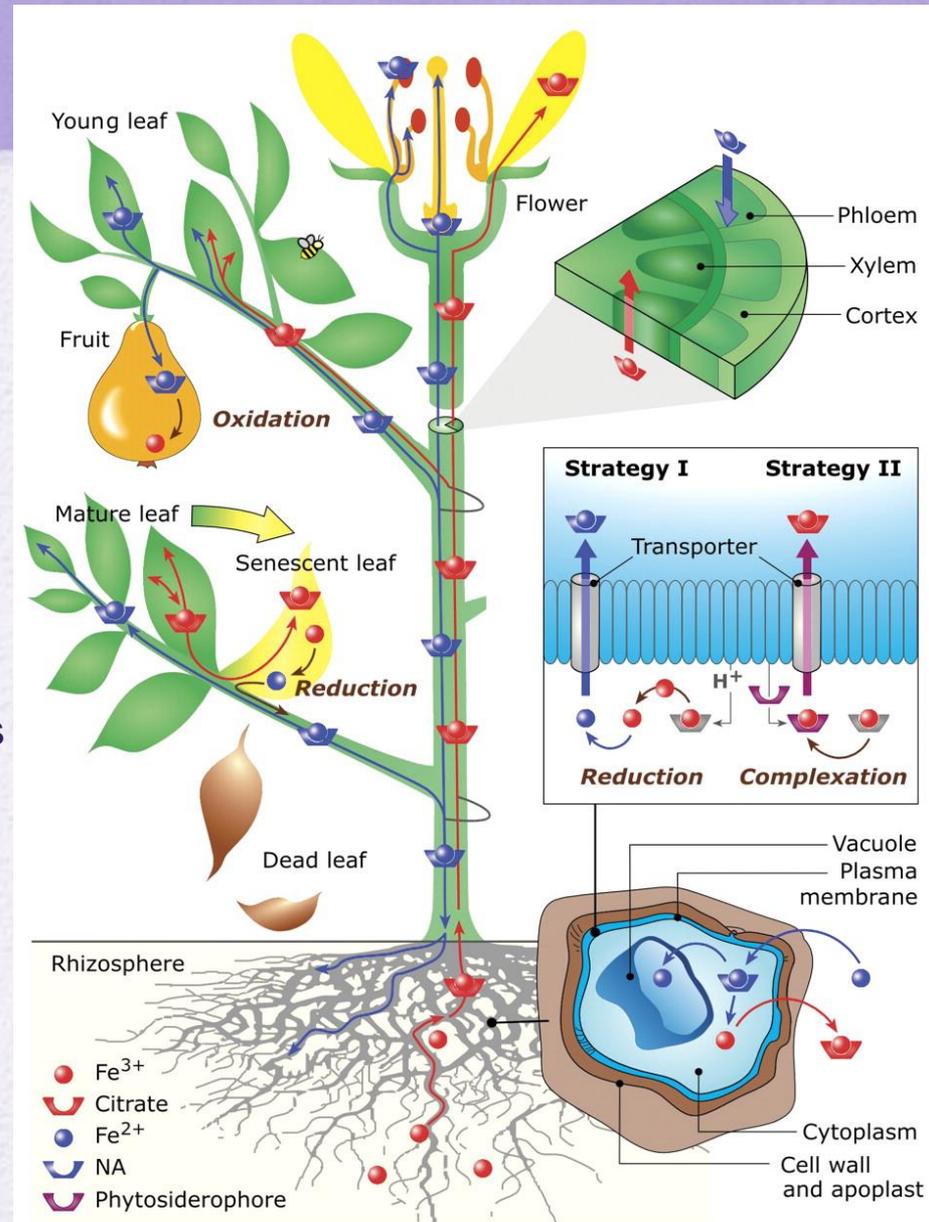
- $\beta$ -glucosidase: phenol oxidase ratio (BG:PPO).
- Cellulose vs lignin degrading enzymes = different pools of carbon in the soil.
- Represents the long-term capacity of the microbial community to continue to metabolize recalcitrant C sources.



# Q2: Microbial Activities:

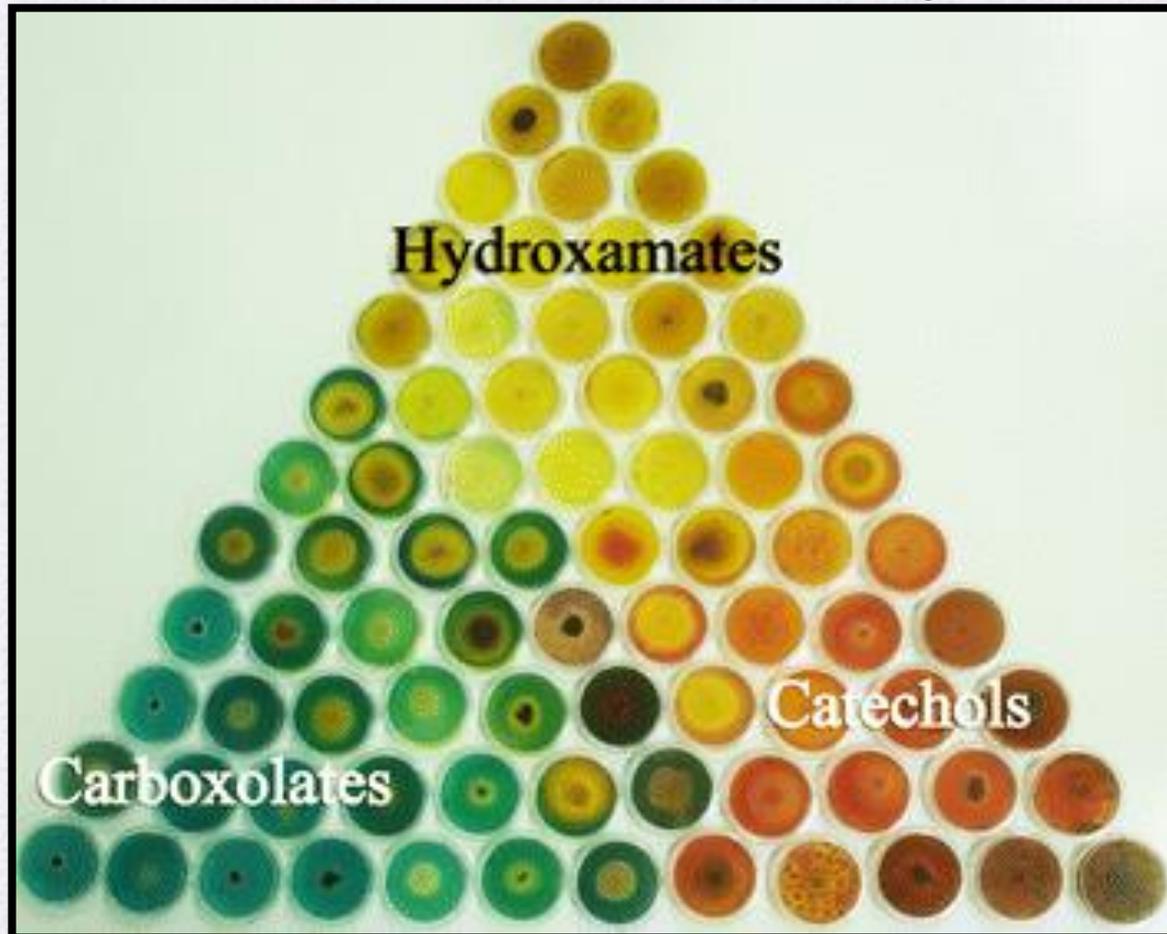
## Siderophores

- Siderophores =
  - Ligands/Chelators
  - Signals
  - Nutritional enhancement
    - Microbes exude dozens of different kinds



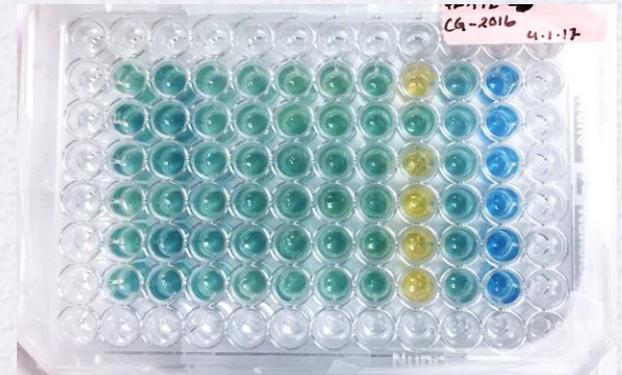
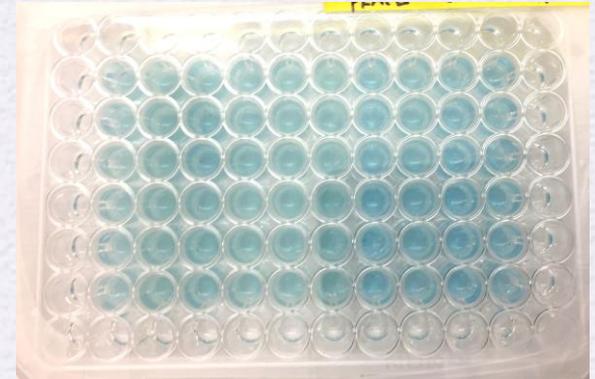
# Q2: Microbial Activities: Siderophores

- Traditional CAS Assay



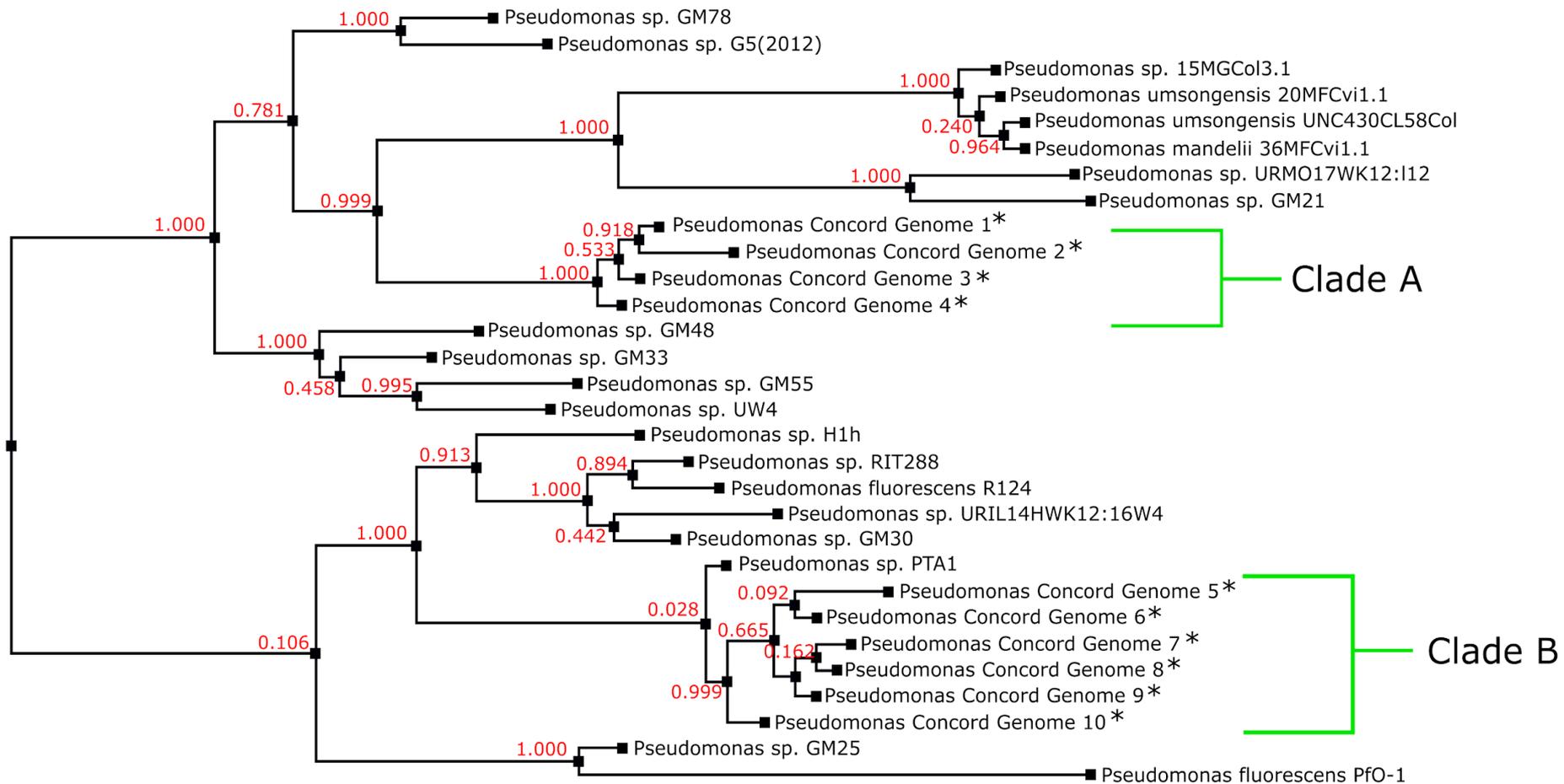
# Q2: Methods Development

- Microtiter assay for rapid screening of many soil samples (many vines, whole vineyards) simultaneously
- 6 replicates of each sample
- Positive and negative controls
- Results in 24-48 hrs
- Quantitative on plate-reader



Lewis, R., Islam, A., Dilla-Ermita, J.C., Hulbert, S.H., and T. S. Sullivan. High-throughput siderophore screening from environmental samples: plant tissues, bulk soils, and rhizosphere soils. *Journal of Visualized Experiments* (In Press: <https://www.jove.com/in-press>).

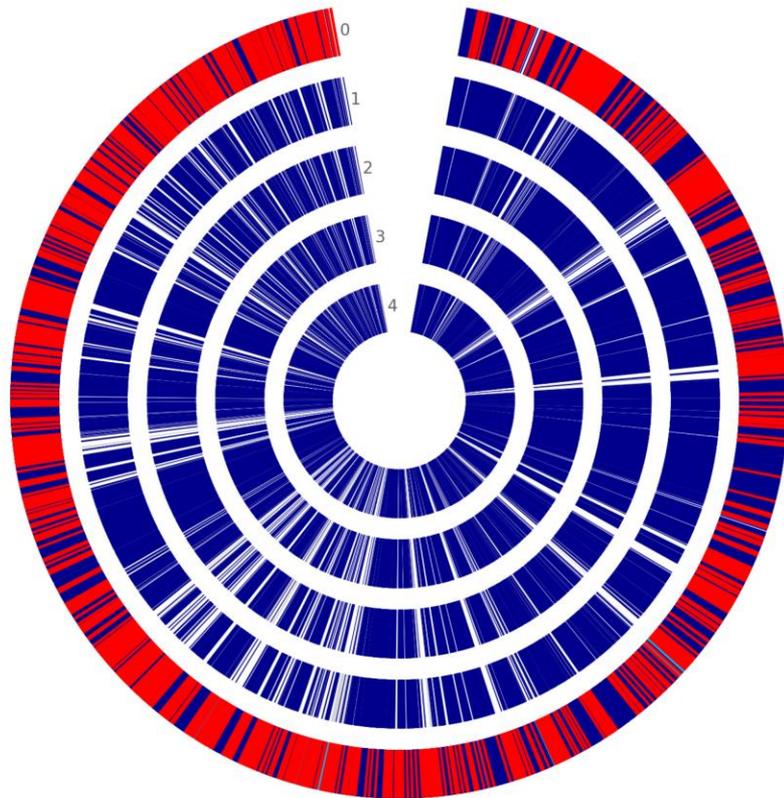
# Q2: Organisms From Microtiter Wells



Lewis, R., Opdahl, L., Islam, A., Davenport, J., and T. S. Sullivan. Comparative genomics, siderophore production, and iron scavenging potential of root zone soil bacteria isolated from 'Concord' grape vineyards. *Microbial Ecology* (In revision).

# Q2: Organisms From Microtiter Wells

A



*Pseudomonas* sp. GM78

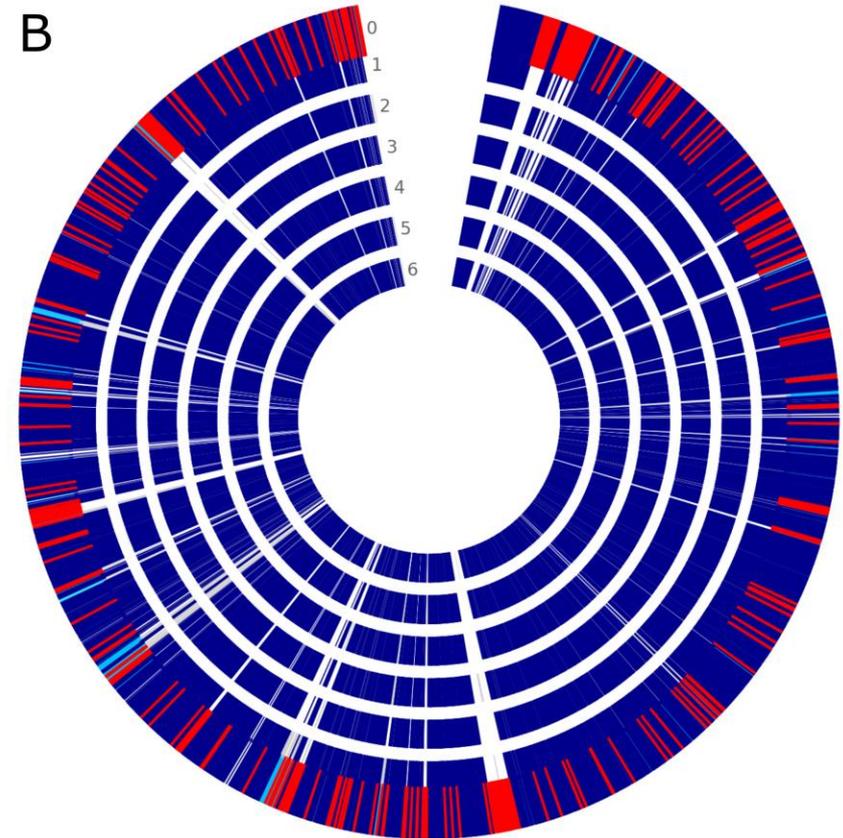
■ Base Singletons  
■ Non-core  
■ Core

Pangenome

■ Non-core  
■ Core

0 *Pseudomonas* sp. GM78  
 1 *Pseudomonas* Concord Genome 4  
 2 *Pseudomonas* Concord Genome 3  
 3 *Pseudomonas* Concord Genome 1  
 4 *Pseudomonas* Concord Genome 2

B



*Pseudomonas* sp. PTA1

■ Base Singletons  
■ Non-core  
■ Core

Pangenome

■ Non-core  
■ Core

0 *Pseudomonas* sp. PTA1  
 1 *Pseudomonas* Concord Genome 9  
 2 *Pseudomonas* Concord Genome 10  
 3 *Pseudomonas* Concord Genome 6  
 4 *Pseudomonas* Concord Genome 8  
 5 *Pseudomonas* Concord Genome 7  
 6 *Pseudomonas* Concord Genome 5

# Q2: Summary

- Microbial activities clearly enhanced under cover cropping.
  - Indicators of increased soil health directly under cover crop.
- Enhanced activity does not reduce chlorosis.
  - Short-term study; long-term soil stewardship?
- Siderophore “cheating” possible competing with chlorotic vines for essential Fe at a time when vine most vulnerable.

# Conclusions

- Root exudates of chlorotic vines shift bacterial community composition and function.
- Potentially allowing open niches for deleterious organisms to supplant plant-growth promoting bacteria.
- Long-term soil stewardship, enhancing soil health, likely leads to healthier vineyards overall.

# Future Directions

- Definitive link between siderophore production and grapevine health, yield, and vigor.
- Greater characterization of grapevine root exudates and how they drive soil health.
- Long-term understanding of the process of microbial community change throughout the health/life of the vine.

# Many Thanks...

- WSU Crop and Soil Sciences
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