

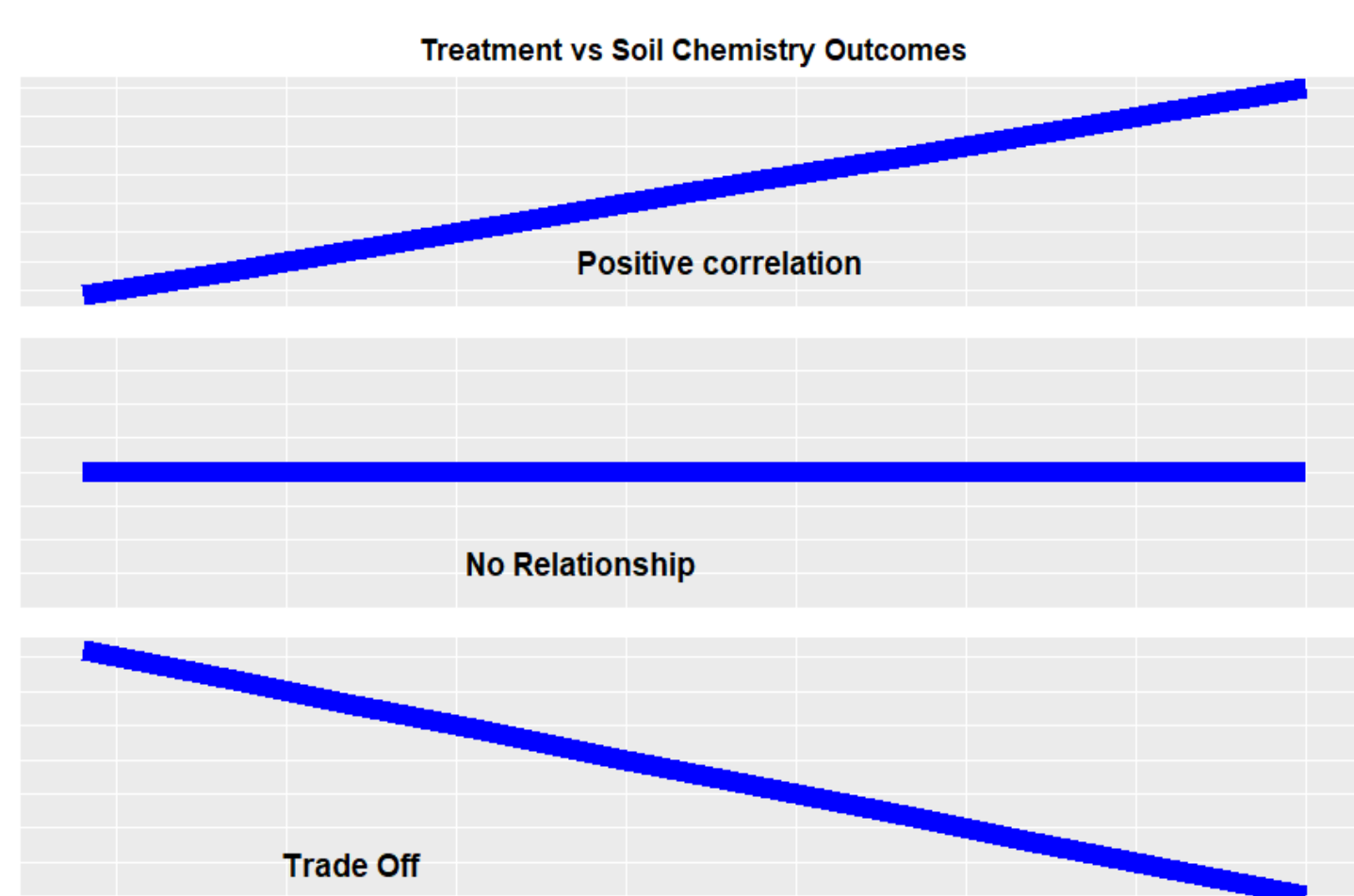
Introduction

Mesorhizobium soil bacteria participate in a mutualism with legumes species. The rhizobium exchange nitrogen fixed from the environment, for carbon sugars the host provides. This mutualism is used in the farming of human food crops to supplement industrial fertilizer. Biotic conditions that affect this mutualism include growth-inhibiting heavy metals and exposure to ultraviolet radiation. Serpentine soil high in concentrations of heavy metals, such as nickel, tend to have low-density growth with short plants. In industrial farmlands where water is scarce waste-water irrigation is used.

Heavy metals in the water column are captured in this wastewater are sprayed all over the crops creating a gradient of serpentine soil. Because the heavy metal does not degrade it remains present in the soil crop after crop.



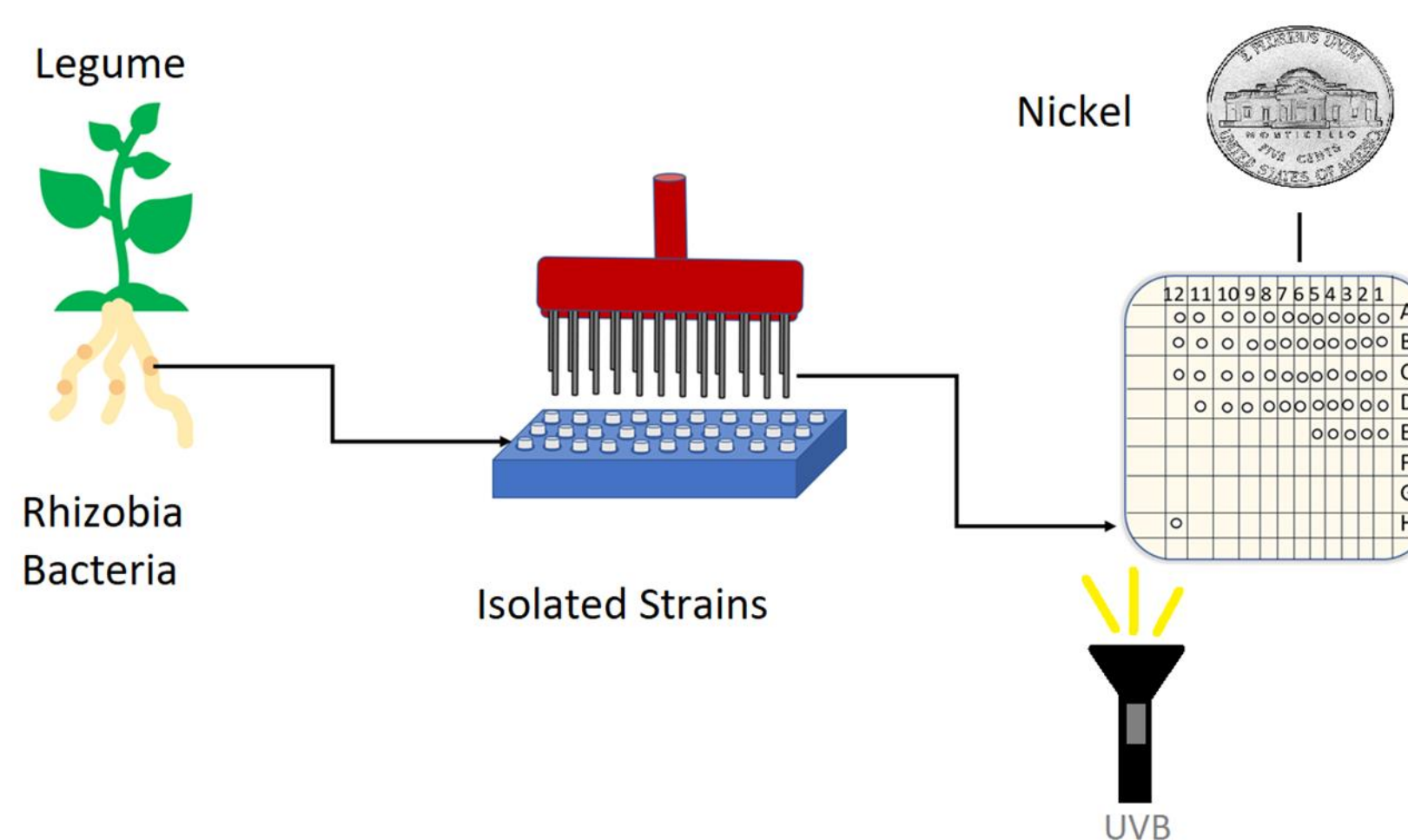
Some legumes can thrive in nickel-rich serpentine soils where other plants cannot. This may be a result of local adaptation for heavy metal and UV radiation tolerance by the *Mesorhizobium* partner. If we determine local adaptation, we can select specific *Mesorhizobium* strains tolerant to heavy metal to use in the bioremediation of polluted industrial farmlands. Local adaptation can be shown for heavy metal by having the same nickel minimum inhibitory concentration versus soil concentration across evolutionary time indicated by spanning multiple clades in the phylogenetic tree. The possible outcomes of comparing treatment vs soil chemistry is represented in the included graph.



Objectives

The objective of the study is to identify if strains in heavy metal rich environments with increased ultraviolet radiation have higher tolerance to nickel and ultraviolet radiation than those found elsewhere. Additionally, we wanted to identify the relationship between UV and nickel tolerance and use it in combination with a phylogenetic tree to determine if these traits are a result of local adaptation.

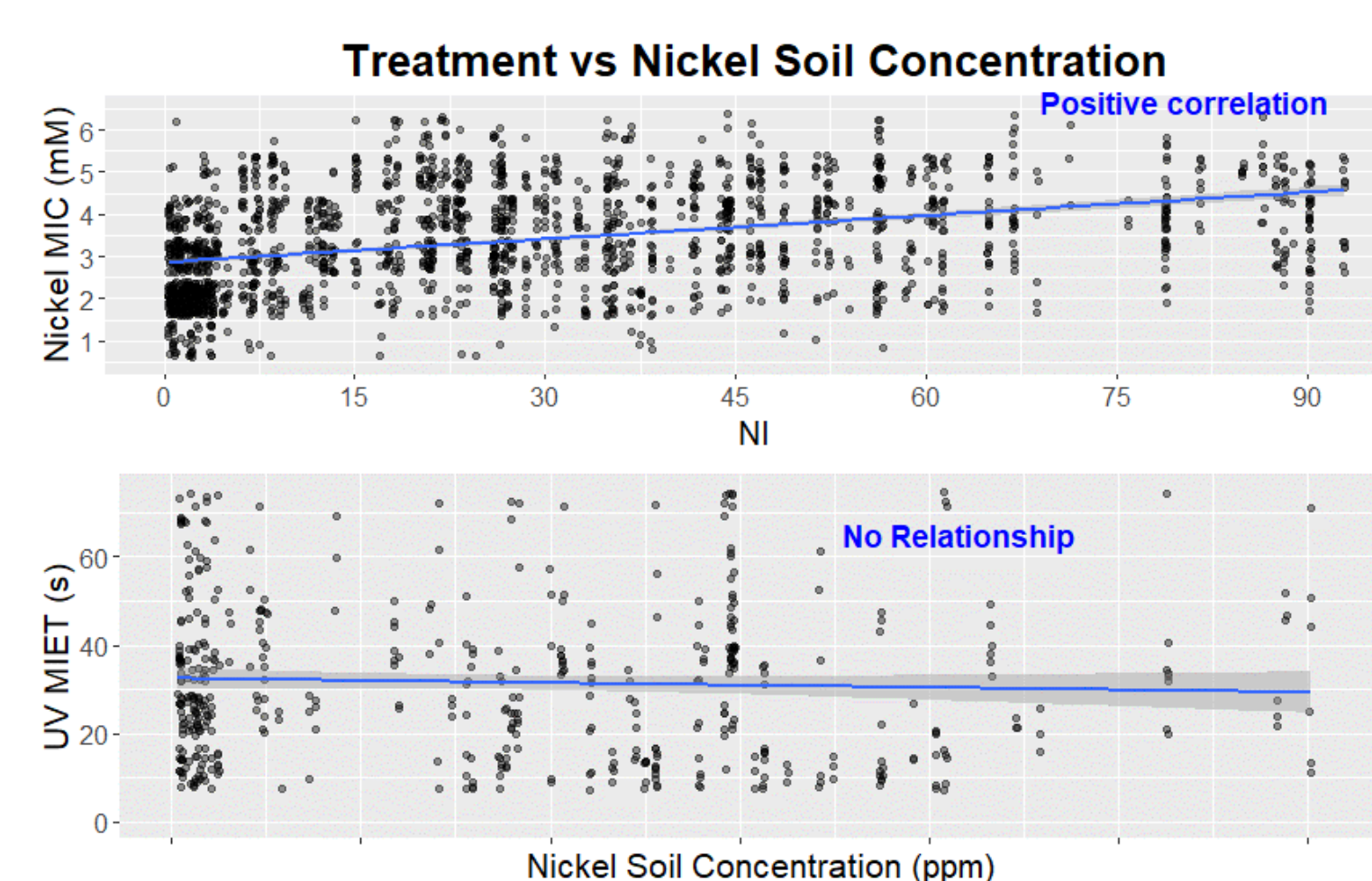
Methods



We grew each of our 770 strains on nutrient enriched media agar plates. The nickel treatment consisted of adding nickel chloride to the agar in one millimolar (mM) increments ranging from zero to five mM. In the UV treatment, the strains on nutrient agar plates were then exposed to UV-b light for up to 60 seconds. The presence of a colony for each strain was recorded 8 days after treatment and used to determine relative nickel and UV tolerance. We used linear modeling to determine the relationship between the treatment and soil chemistry, as well as the relationship between nickel and UV tolerance.

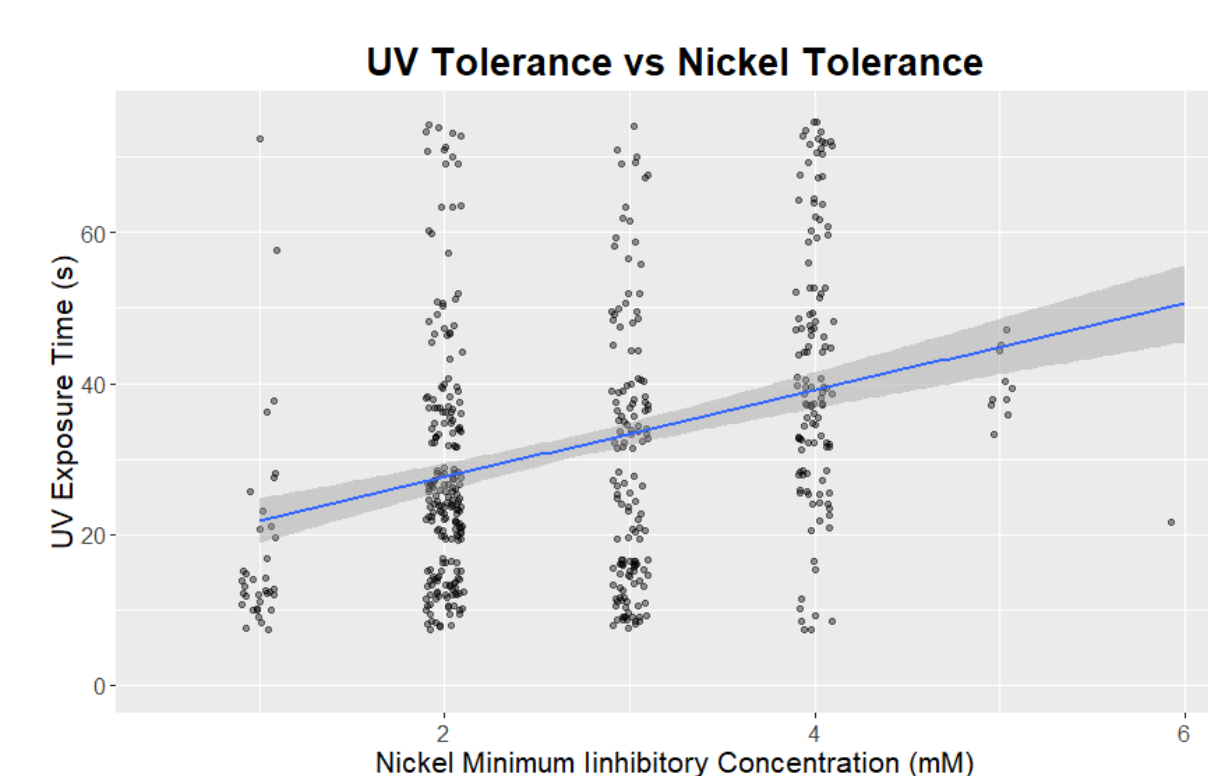
Results

Is there tolerance for nickel and UV exposure?



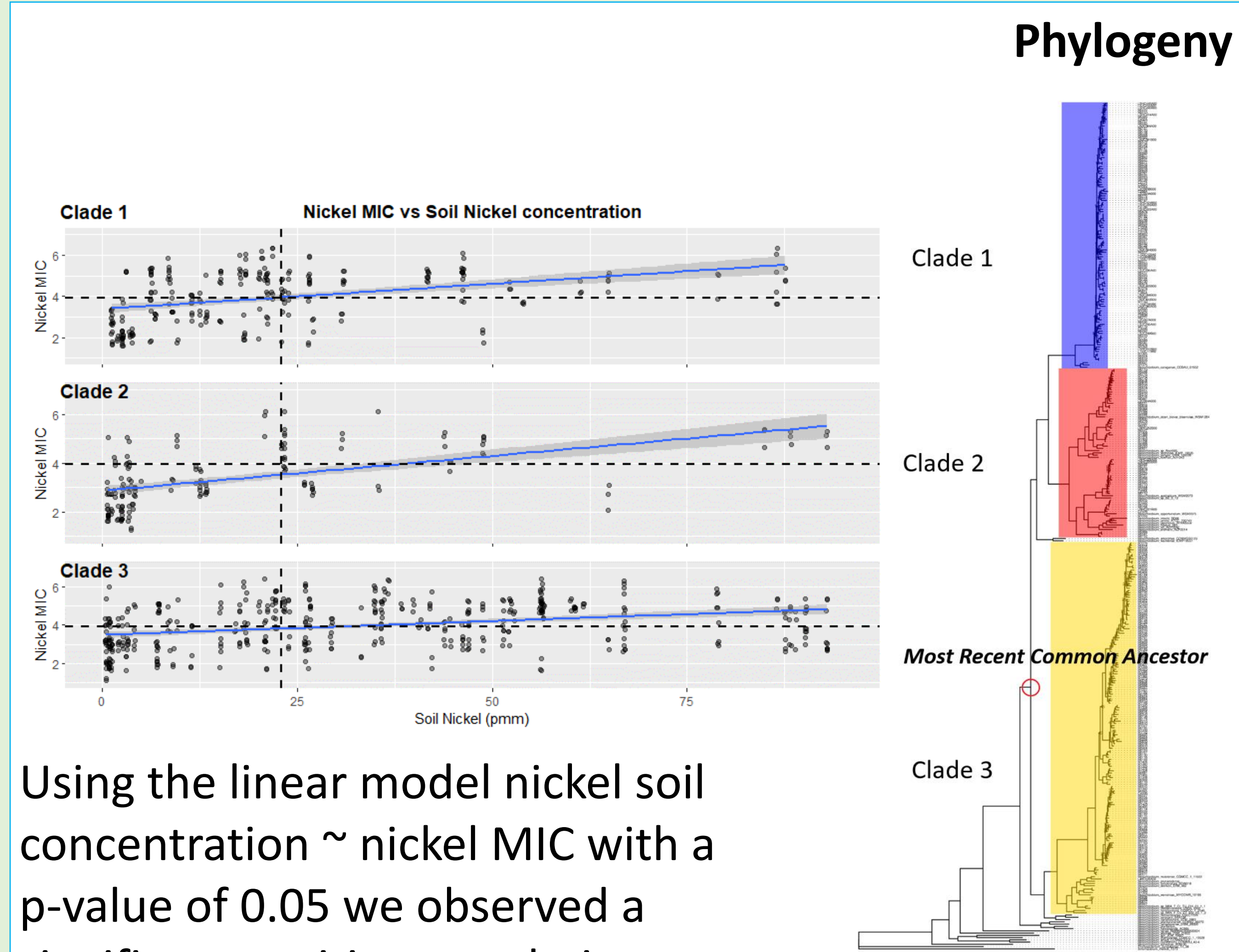
- Using the linear model nickel soil concentration \sim nickel minimum inhibitory concentration (MIC) with a p-value of 0.05 we observed a statistically significant positive correlation (p-value $<$ 0.05).
- Using the linear model nickel soil concentration \sim UV minimum inhibitory exposure time (MIET) with a p-value of 0.05 we did not observe a statistically significant correlation.

How are nickel and UV tolerance related?



- Using the linear model UV MIET \sim nickel MIC with a p-value of 0.05 we observed a statistically significant positive correlation (p-value $<$ 0.05).

Results continued



Using the linear model nickel soil concentration \sim nickel MIC with a p-value of 0.05 we observed a significant positive correlation across clades 1,2, and 3.

Conclusions

We found a positive correlation between nickel minimum inhibitory concentration and soil nickel concentration, meaning that as soil nickel increases so does the *Mesorhizobium's* nickel tolerance increases. With this behavior being constant across all 3 of our experiment's phylogeny we have enough to determine local adaptation across evolutionary time. UV tolerance was not correlated with soil chemistry but did show a positive correlation with nickel tolerance. This means as nickel tolerance increase so does UV tolerance, which is what we expected to see. With more UV exposure data incorporated into the phylogeny we could make a conclusion about local adaptation but as of now we can only show correlation. With heavy metal adaptation being shown we can use nickel soil concentration to choose an appropriate *Mesorhizobium* strain to help in bioremediation of wastewater irrigated farmlands.

Continued Research

- I want to expand my strain sample size.
- I want to incorporate the UV tolerance comparison to the phylogeny mapping.
- Perhaps expand this project to include an antibiotic treatment.

Acknowledgments

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