

## Introduction

**How does Tobacco mosaic virus impact the biodiversity of *Nicotiana benthamiana* plant?**

**Defense Pathways of Plants**

- Organic compounds such as Ethylene, Salicylic Acid, and Jasmonic Acid
- Survival of organotrophs and stimulates plant defenses
- Streptomyces* acts as a biofertilizer(13)

**GAP IN KNOWLEDGE**

- Comparisons of the microbe communities between parts of the plant's phyllosphere
- Coffea arabica* plant indicates that after viral incubation, species richness and calcium levels (11) decreases
- Beneficial microbes CAN become antagonistic(12)

**Hypothesis:**  
The presence of TMV will decrease the biodiversity of microbes.

**Structure of TMV virus(9)**

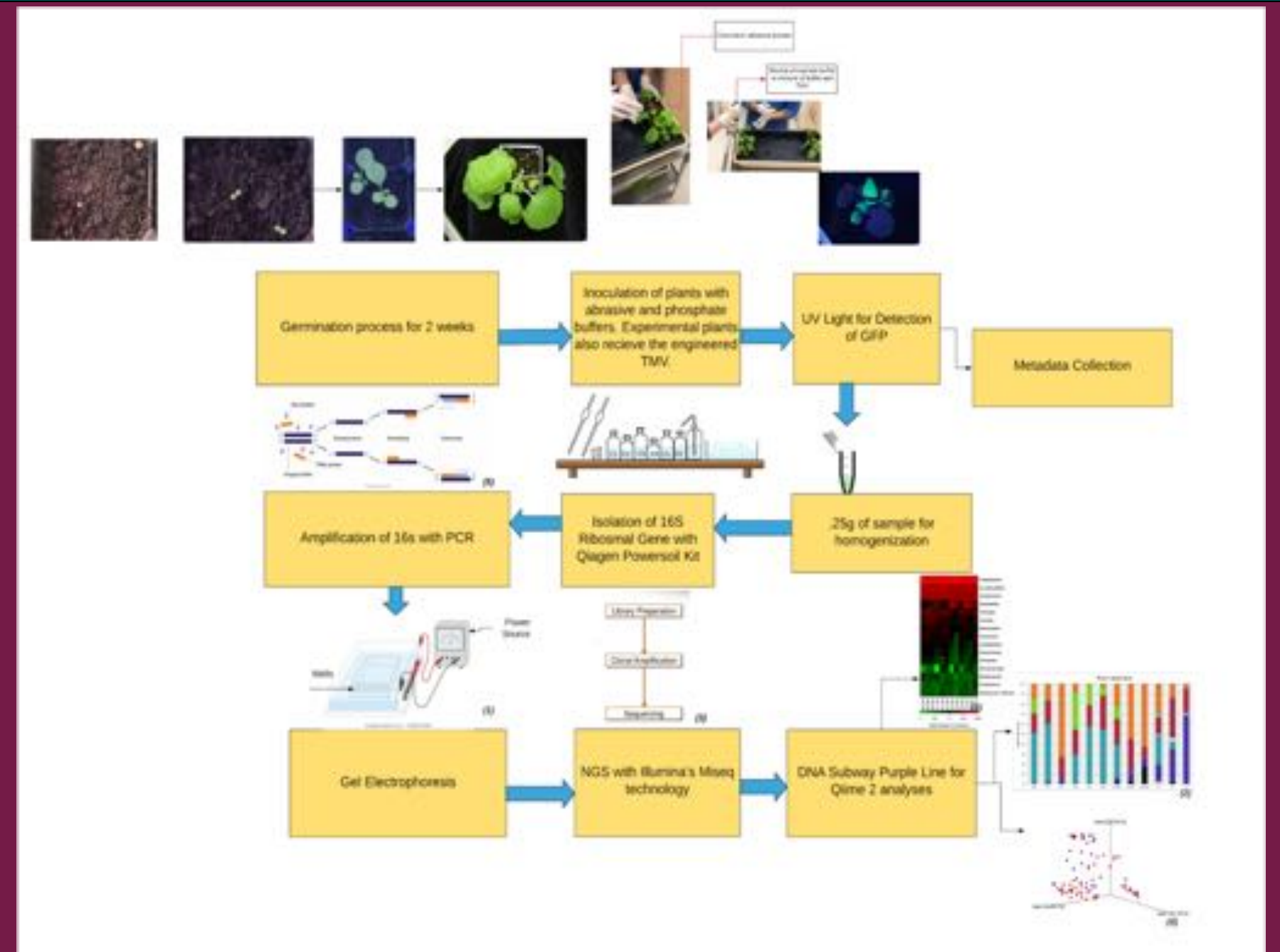
- Decline in tobacco yields
- All ranges of Solanaceous crops transports virus to the plant's mechanical wounds(10)

A phyllospheric analysis indicated how TMV impacts the microbial diversity in the plants, especially due to how microbes can have dual characteristics. In addition, with a biochemical analysis, plant management plans can be established in order to prevent the spread of TMV to one crop to a neighboring crop.

## Aims

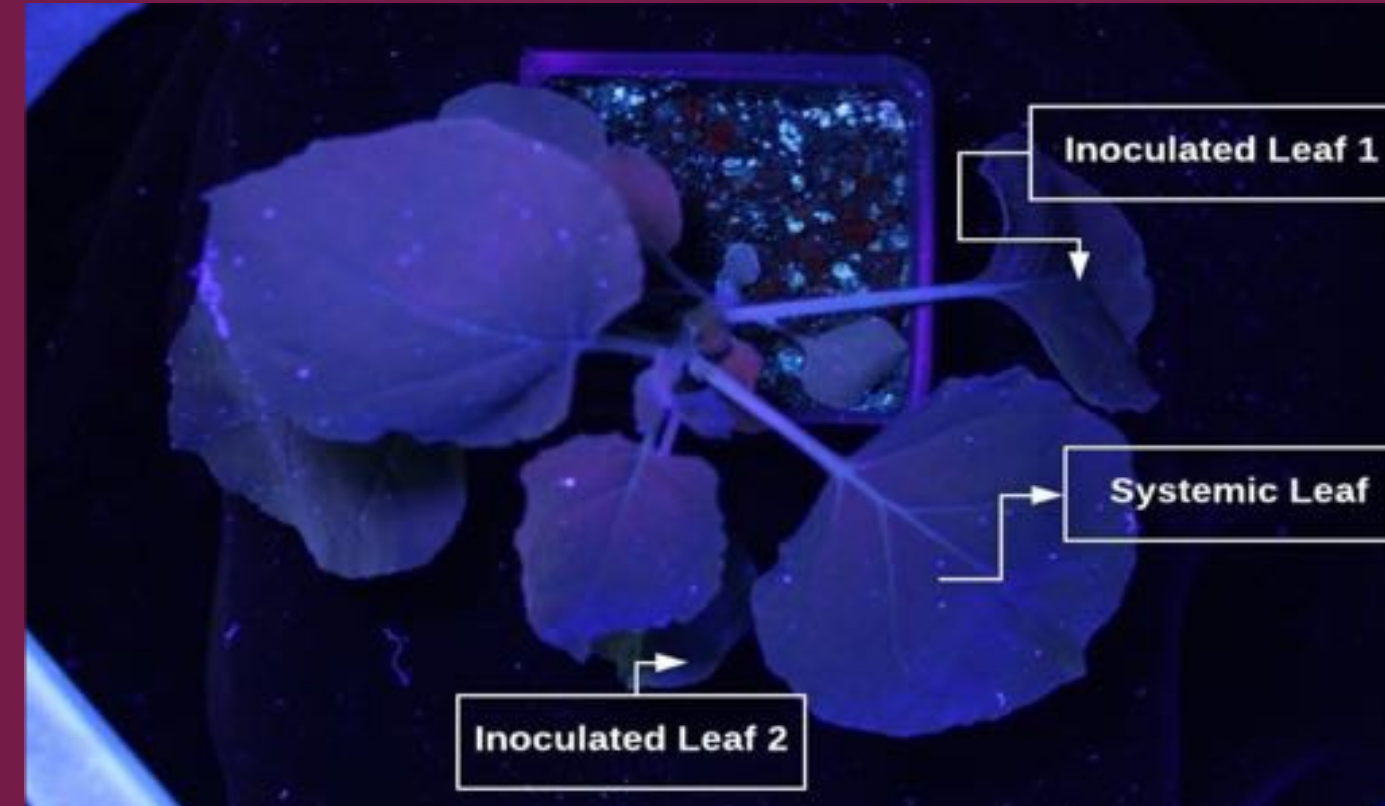
- To expose TMV to healthy *Nicotiana benthamiana* plants through mechanical inoculation on leaves.
- To conduct microbial analyses on the plant's leaves and soil with and without TMV.

## Methodology



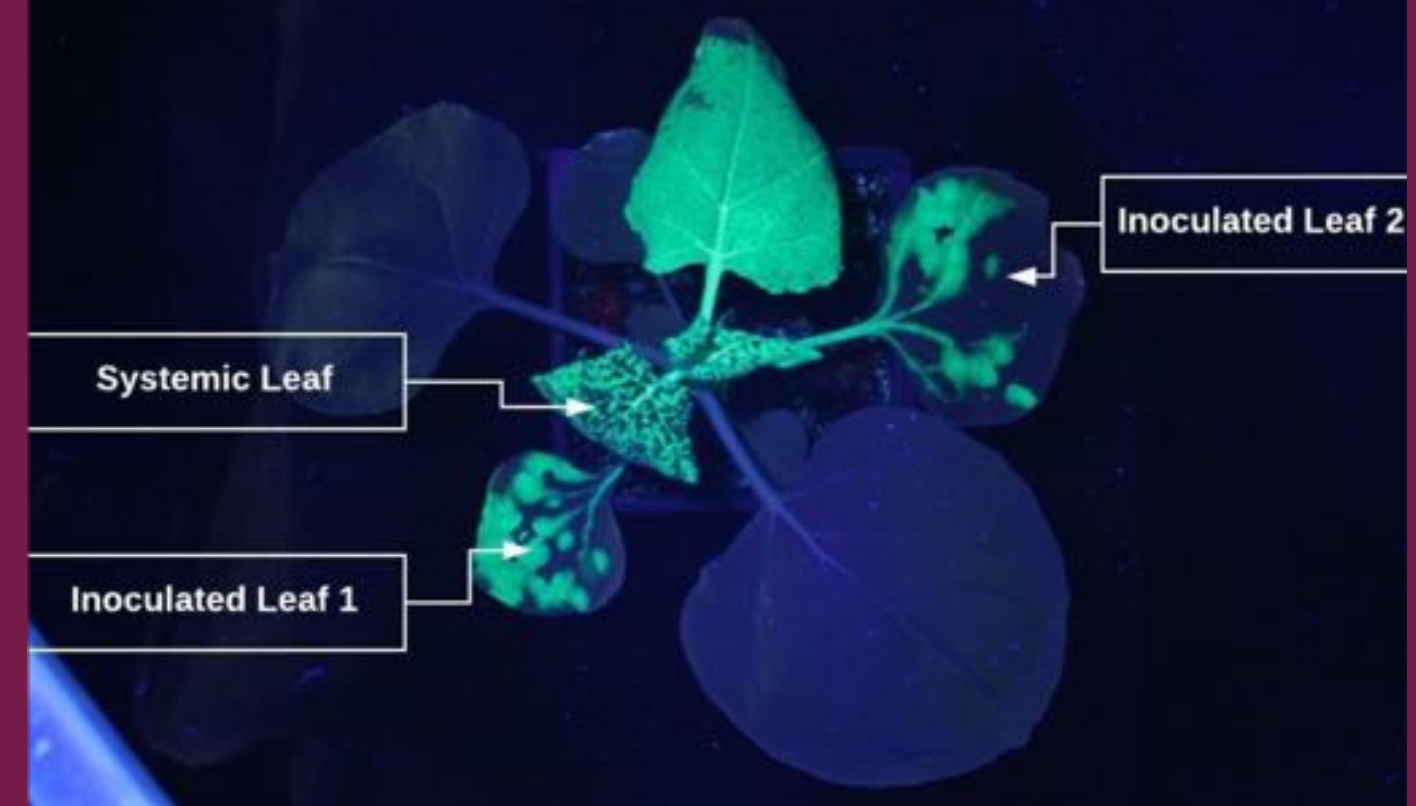
## Results

**Figure 1: Mock Inoculated Plant under UV lights**



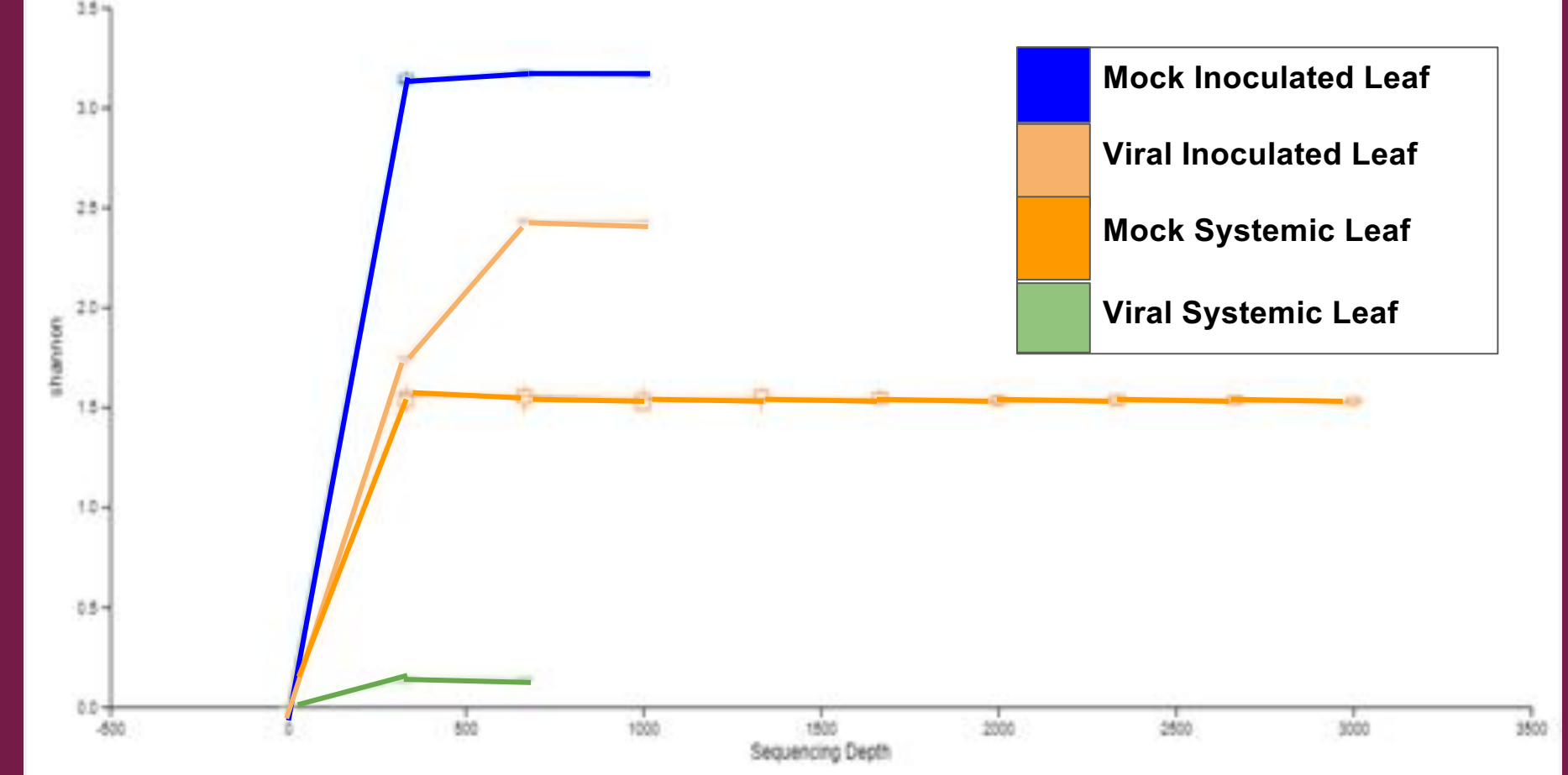
Three plants were mock inoculated with a phosphate buffer and abrasive. It is evident that there is no exogenous signs of stress because there's no presence of virus.

**Figure 2: Viral Inoculated under UV Lights**



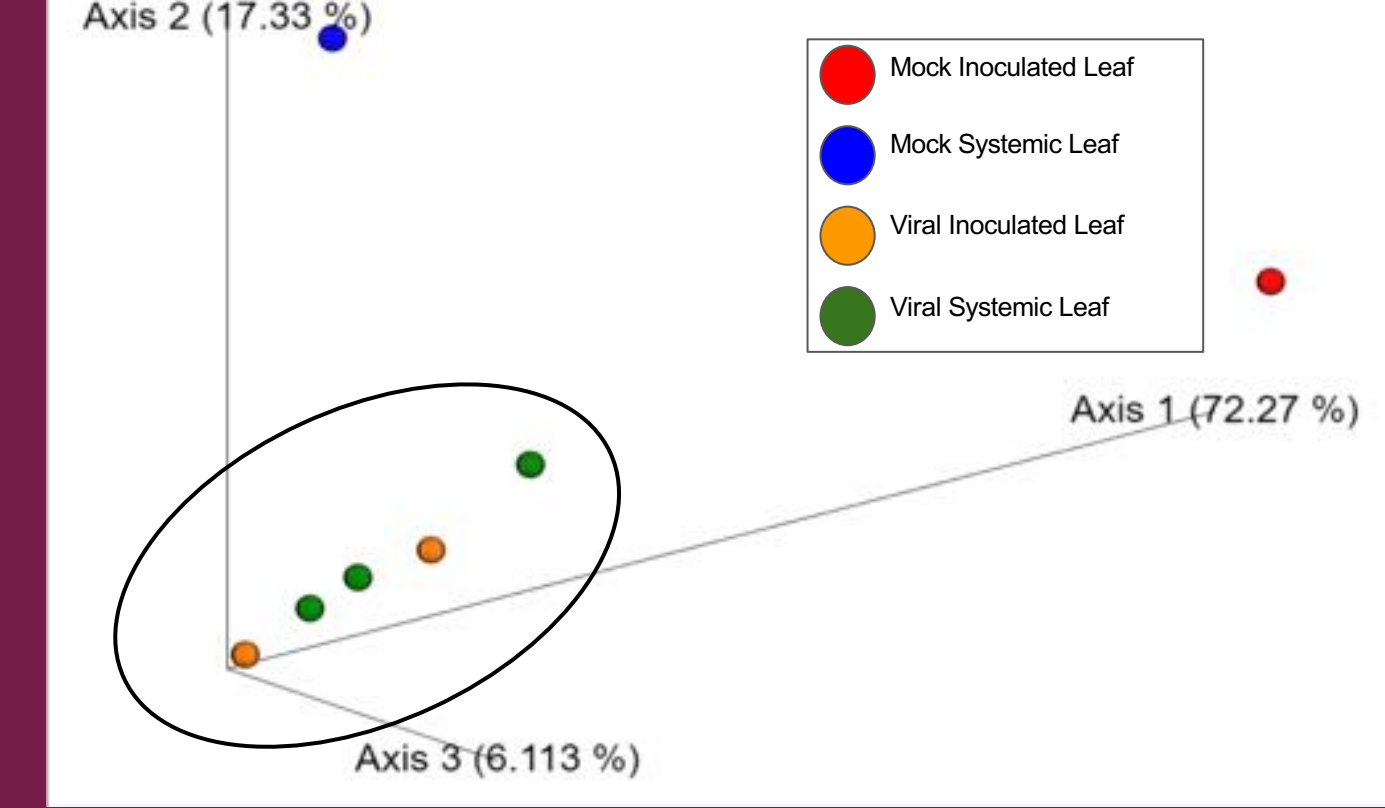
The additional three plants were used as viral samples; inoculated with phosphate buffer, abrasive and fluorescent TMV. The fluorescence indicates the presence of viral colonies.

**Figure 3: Species Richness in Organs of Plants**



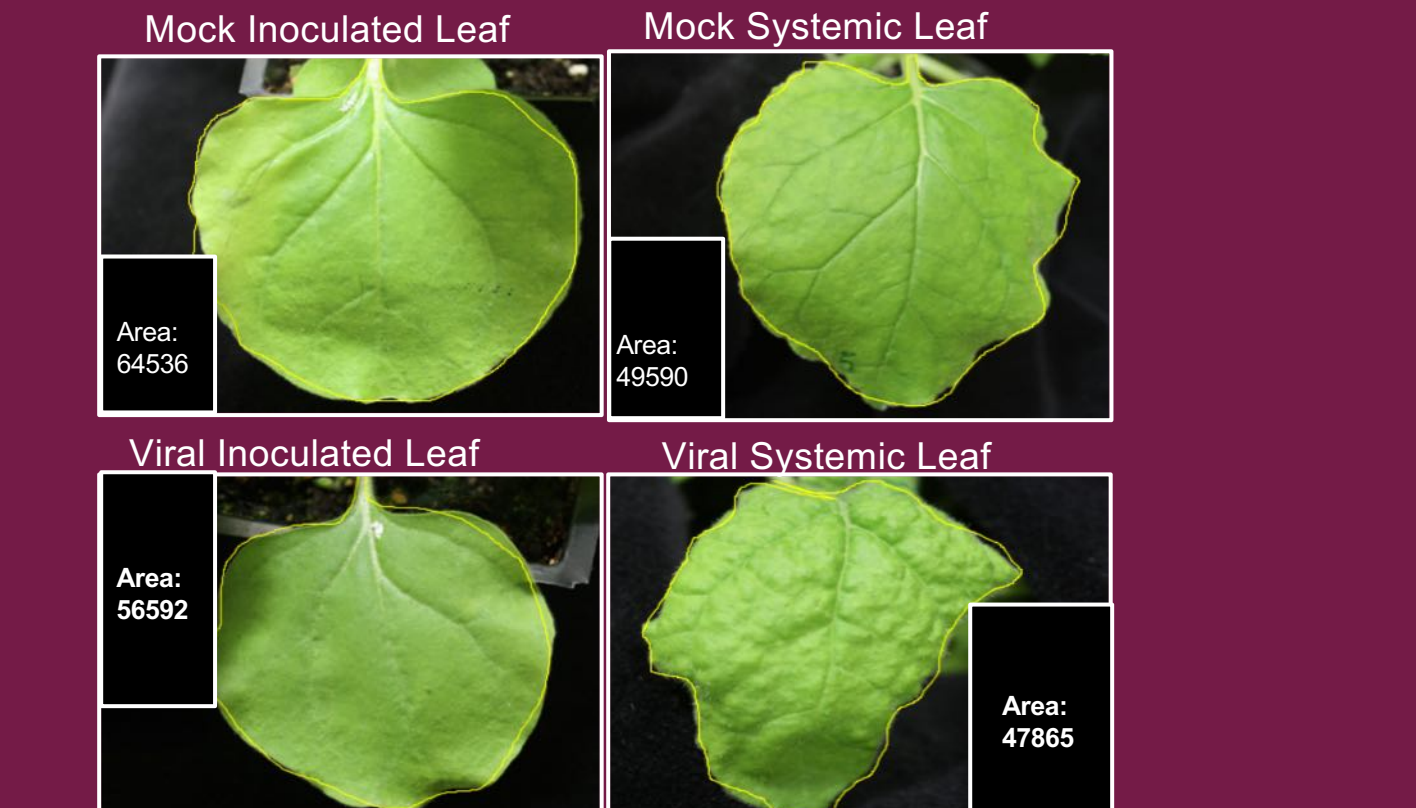
The rarefaction plot displays the species richness between the plants organs. The site with the highest species richness was in mock inoculated leaves followed by the viral inoculated leaves. This similar pattern was seen in the mock systemic leaves where there was the highest species richness compared to the viral systemic leaves.

**Figure 4: Comparisons between Plant Organs**



The comparison between the plant organs are indicated by the distance between each sample. There is clustering between all 5 of the viral samples, further indicating that the samples have similar species presence.

**Figure 5: Surface Area of Mock and Viral Leaves**



Both mock samples have a higher area than viral samples. However, the mock systemic has a slightly higher area compared to the viral systemic leaf.

## Discussion

**Trends in Microbial Presence**

**Research Question-** How does TMV impact the biodiversity of *Nicotiana benthamiana*?

**Hypothesis-** The presence of microbes will decrease due to RNA virus.

**Species Richness**

- As illustrated by the figure 3, both mock inoculated and systemic leaves had higher species richness than the viral inoculated and systemic leaves.
- The rarefaction further supports the hypothesis regarding that species richness was highest in plant organs where there was no presence of antagonistic TMV.
- Previous research on subtropical trees indicates that leaf size may be directly related to species richness (18).
- The systemic leaves normally have lower species richness because they are young compared to the matured inoculated leaves
- In addition, figure 5 is indicating that antagonistic biotic stress may decrease the growth rate of the systemic leaf. Consequently, resulting in lower species richness. This could be the case because the mock systemic has a slightly higher area than the viral systemic leaf.

**Microbial Variation**

- According to previous research regarding flue cured tobacco, specific taxa may not have been generated because mitochondrial and chloroplast DNA contaminate sequences (15)
- In addition, defense mechanism such as **Systemic Acquired Reduction** played a crucial role endogenously to increase microbe presence in the viral inoculated leaves and later the viral systemic leaves. This was indicated by the clustering of all viral samples in figure 4 and the low species richness of the viral samples compared to the mock samples in figure 3
- Control samples may not be clustering due to small sample size.

## Conclusion

The hypothesis was further supported by figure 3 and 5 which indicated that both mock inoculated organs had higher species richness than the viral organs. In addition, figure 4 illustrates that there is clustering between all viral samples which indicates that the samples have similar taxa presence due to the defense mechanisms such as induced systemic acquired reduction repairing and defending the plant from biotic stress.

## Future Implications

- Increasing sample size for this study would allow for stronger statistical significance especially in the pcOA plot.
- Shifting to analysis of the rhizosphere, including the soil and roots
- Through Jupyter Notebook, the mitochondrial and chloroplast DNA can be filtered out.
- Getting specific to a family or phylum of the microbes with the use of 8F primer
- Analyzing both chemical defense mechanisms and toxins such as salicylic acid, jasmonic acid, ethylene, and HC Toxin through infrared microspectroscopy