What microbes are found in watermelon (*Citrullus lanatus)* flowers and seeds?

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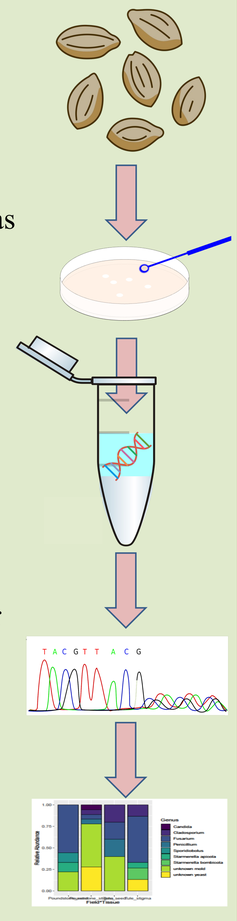
Background

- Seed microbes contribute to young plant health (1) and microbial community formation (2-3).
- Floral stigmas are a potential source of microbes for seeds (4).
- The overlap of stigma and seed microbes is understudied (5).
- Watermelon *(Citrullus lanatus)* is a good model plant because of its large flowers and seeds (6), and it has yet to be surveyed for microbes in detail (7).
- **Research questions:**
  - **Which microbes are shared between stigmas and seeds in watermelon *(Citrullus lanatus)?***
  - **Do these microbial communities vary between plant tissues and fields?**

Methods

- Collect floral stigmas and seeds from two fields (Poundstone and Tule)
- Isolate bacteria and fungi from stigmas and seeds in culture
- Extract and amplify microbial DNA from cultures for Sanger sequencing
- Identify microbes by querying Sanger sequences against DNA databases
- Describe microbial community composition



Results

20% of stigma bacteria (Fig. 1A) and 77.8% of stigma fungi (Fig. 1B) were also found in seeds. Bacterial richness did not vary by plant tissue (ANOVA; F=1.83, p=0.186) or source field (ANOVA; F=1.07, p=0.309), but fungal richness was higher in stigmas than seeds (ANOVA; F=6.36, p=0.0172).

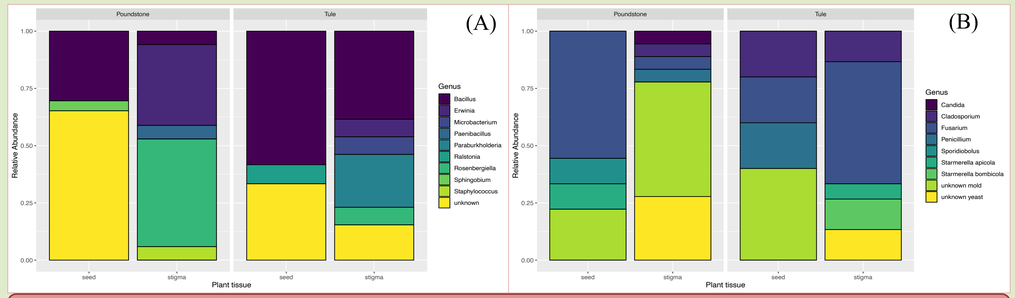


Figure 1. Taxonomic composition of culturable bacterial (A) and fungal (B) communities of *C. lanatus* flowers and seeds. Relative abundance was calculated as the number of isolated identified to a particular genus over the total number of isolates from that tissue*field source.

Microbial isolation frequencies from seeds were very low, and there was no significant difference in bacterial (Fig. 2A; T-test: t=-0.017, p=0.87) and fungal (Fig. 2B; T-test: t=-0.67, p=0.53) frequencies between source fields.

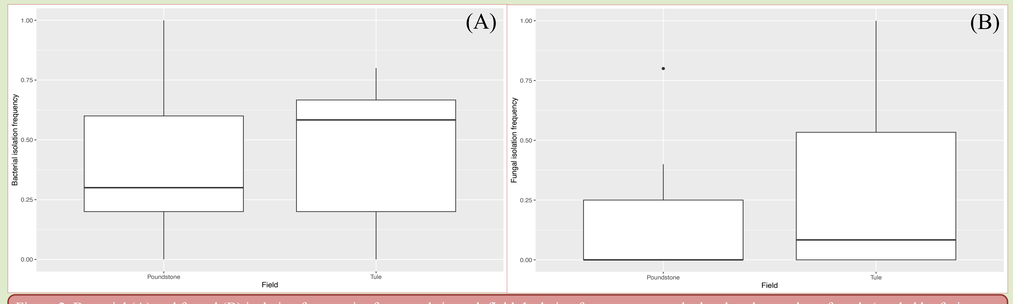


Figure 2. Bacterial (A) and fungal (B) isolation frequencies from seeds in each field. Isolation frequency was calculated as the number of seeds (pooled by fruit slice) that had a microbe over the total number of fruit slices.

Bacterial communities varied by field (Fig. 3A; PerMANOVA: F=2.99, p=0.022) and plant tissue (Fig. 3A; PerMANOVA: F=4.58, p=0.003), and fungal communities also varied by field (Fig. 3B; PerMANOVA: F=7.52, p=0.001).

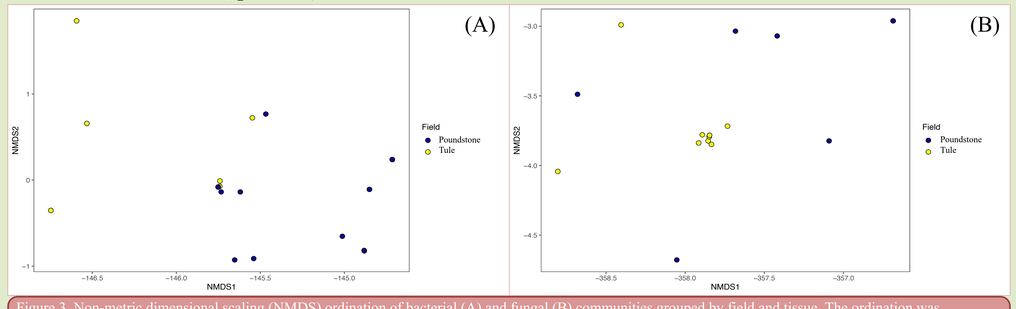


Figure 3. Non-metric dimensional scaling (NMDS) ordination of bacterial (A) and fungal (B) communities grouped by field and tissue. The ordination was calculated from a Jaccard (i.e., presence/absence) dissimilarity matrix of bacterial and fungal communities from flowers and seeds. Points that are closer together represent communities that are similar to each other, while points that are far apart represent communities that are different from each other.

Conclusions

- **The most common microbes found across plant tissues and fields were *Bacillus* and *Fusarium,* which have been found in seeds of other plants (6-8).**
- **There was lower microbial presence and richness in seeds compared to flowers, which is consistent with previous research in other plants (6) and aligned with the Primary Symbiont Hypothesis (9).**
- **There was some variation in microbial community richness and composition between fields, which could be due to crop variety, environmental conditions or both.**

Future Work

- Test which microbes in our culture collection move from flowers to seeds in inoculation experiments.
- Test for antagonistic interactions between *Bacillus* and watermelon seed pathogens in culture.
- Test if seed inoculation with our culture collection affects seed/seedling survival.

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