Influence of bacterial inoculum density on host fitness

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Introduction

- Families of coreoid and lygaeoid bugs acquire their symbiont from the environment.
- *Leptoglossus phyllopus is a* common polyphagous agricultural pest found in North and South America, and primarily targets citrus and tomatoes.
- *Callaberonia is a bacterial symbiont* crucial for *L. phyllopus* survival and development [1, 2]

Research Question: How does bacterial inoculum density affect host fitness?

Hypothesis: L. phyllopus infected with higher concentration of the Caballeronia will exhibit faster development and better fitnes

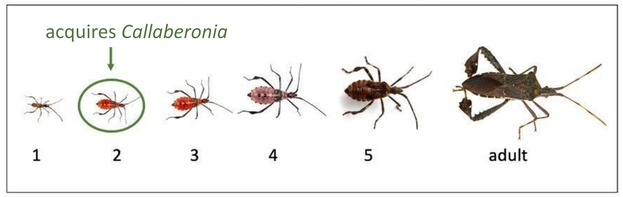

Figure 1. Development stages of *L. phyllopus*
The photo illustrate each stage of development of *L. phyllopus* and when they acquires their symbiont.

Methods

- *Caballeronia* isolate, LEP1A1, were cultured in shaking incubator at 28°C, at 280 rpm
- 2nd instar nymphs were infected for 24 hours with three different concentration of *Caballeronia* : 1 x 10^4 cells/mL (low), 1 x 10^6 cell/mL (Medium), and 2 x 10^7 cell/mL (High).
- Nine replicate trials were conducted for each of the various concentration
- Once *L. phyllopus* were infected, the development time to adulthood and the mass at adulthood were measured

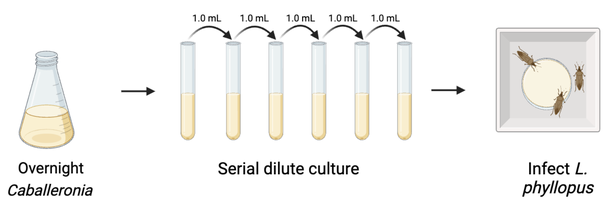

Figure 2. Illustration of infection box method.

Results

- There is no correlation between *L. phyllopus* development time (Fig.3) and adult mass (Fig. 4) amongst the three treatment, therefore inoculum density could not be associated with host fitness.

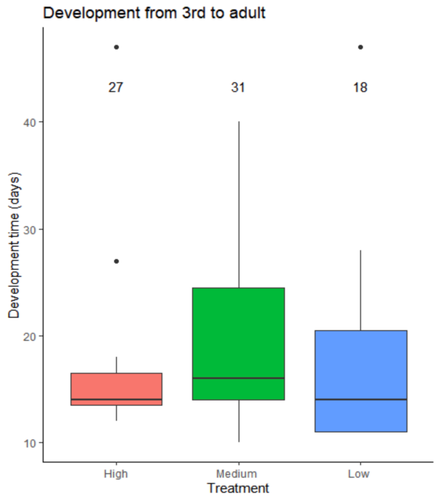

Figure 3. The time it took for each experimental group of *L. phyllopus, infected with Caballeronia,* to reach adulthood. The data was gathered every 1-2 days to monitor the amount of days it took for each *L. phyllopus* from reach 3rd instar to adulthood. (P = 0.6414)

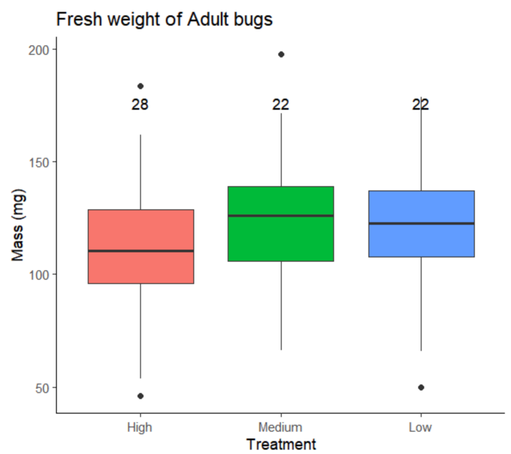

Figure 4. The mass of the three experimental group of *L. phyllopus* infected with *Caballeronia* were measured once they have reached adulthood (P = 0.5975)

Discussion

- The p-value of the mass distribution and development time for the three concentration of the symbiont suggest that fitness of the host was not significantly affected by the symbiont inoculum density.
- Regardless of the initial inoculum density*, Caballeronia* reached required threshold of density inside the host needed for proper development and growth.
- Understanding the relationship between inoculum *Caballeronia* density and *L. phyllopus* fitness would optimize agriculture yield by creating alternative solution to pesticide usage.

Future Research

- Concentration bacterium treatment should be lower considerably to account for natural exposure, to determine the lowest amount needed for *L. phyllopus* survival
- Further studies need to be conducted to account for the total amount of *Callaberonia* in the wild, in the presence of *L. phyllopus* versus environment that does not contain *L. phyllopus*

References

1. Hunter, M. S., Umanzor, E. F., Kelly, S. E., Whitaker, S. M., & Ravenscraft, A. (2022). Development of common leaf-footed bug pests depends on the presence and identity of their environmentally acquired symbionts. Applied and Environmental Microbiology, 88(5). https://doi.org/10.1128/aem.01778-21

2. Kikuchi, Y., & Fukatsu, T. (2014). Live imaging of symbiosis: spatiotemporal infection dynamics of a GFP-labelled Burkholderia symbiont in the bean bug Riptortus pedestris. Molecular ecology, 23(6), 1445–1456. https://doi.org/10.1111/mec.12479

3. Ohbayashi, T., Futahashi, R., Terashima, M., Barrière, Q., Lamouche, F., Takeshita, K., Meng, X. Y., Mitani, Y., Sone, T., Shigenobu, S., Fukatsu, T., Mergaert, P., & Kikuchi, Y. (2019). Comparative cytology, physiology and transcriptomics of Burkholderia insecticola in symbiosis with the bean bug Riptortus pedestris and in culture. The ISME journal, 13(6), 1469–1483. https://doi.org/10.1038/s41396-019-0361-8

4. 5. Kikuchi, Y., Ohbayashi, T., Jang, S. et al. Burkholderia insecticola triggers midgut closure in the bean bug Riptortus pedestris to prevent secondary bacterial infections of midgut crypts. ISME J 14, 1627–1638 (2020). https://doi.org/10.1038/s41396-020-0633-3

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