# Characterization of the Internal Microbiome of a Lab-Reared Urban Arthropod Pest, the Western Black Widow Spider



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Figure 2. Bar plot of the relative

black widow spiders, filtered for those

minimally identified at the genus level.



## INTRODUCTION

- What mechanisms make organisms thrive in cities?
- Black widow spiders are a medically-important pest of the Western U.S. where they form urban infestations.
- The microbiome of this spider species has been relatively unstudied.
- We predict that lab-reared widows will have highly similar microbiome compositions across the sample batch due to controlled rearing conditions.



## METHODOLOGY

- We selected 4 F2 lineage sub-adult spiders, lab-reared for preliminary analysis.
- · Whole spiders were stored at -80°C, preserved in SSC buffer prior to microbiome sample prep.
- Whole lab-reared spiders were surfaced washed with SSC buffer and homogenized with sterile motorized pestles.
- DNA was extracted using Qiagen's PowerSoil Pro Kit and PCR amplified with bacterial V4 region primers.
- 16s rRNA gene sequencing was performed using an Illumina MiSeq instrument and the data was processed with the QIIME2 bioinformatics tool for microbial community analyses.

#### REFERENCES

[1] Trubl, Patricia and J. C. Johnson. Journal of Arid Environments 163 (2019): 18-25 [2] Sugden, S., Sanderson, D., Ford, K. et al. *Sci Rep* 10, 22207 (2020).
[3] Miles, L. S., Ayoub, N. A., Garb, J. E., Haney, R. A., & Verrelli, B. C. (2020). *Genes*, 11(1), 87. Figure 1. Unfiltered bar plot of the relative frequencies of bacterial taxa in 4 lab-reared frequencies of specific bacterial taxa in 4 lab-reared black widow spiders.

100%

90% -

80% -

70% -

60%

nbau 50%

Relative

40%

30% -

20% -

10% -

100% 80% 60% 509 30% 10% **B**8 **B5** MR03 OLI03 Sample Sample

### ACKNOWLEDGEMENTS

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	Taxonomy: Lowest Known Rank	Approx. Frequency (%)	Functional Characteristics
Figure 1.	k_Bacteria; ?	96%	~~~
	k_Bacteria; p_Firmicutes; ?	2%	~~~
Figure 2.	Blautia	0.88%	Liu et. al. (2021): Probiotic properties
	Bacteriodes	0.45%	Significant gut anaerobic pathogen
	Faecalibacterium prausnitzii	0.38%	Promotes good gut health; biomarker
	Enterococcus	0.16%	Normally gut commensal; linked to infections
	Prevotella copri	0.26%	Enriched in those with Rheumatoid Arthritis
	Coprococcus	0.18%	Butyrate-producing bacterium in feces
	Clostridium clostridioforme	0.16%	~~~
	f_Lachnospiraceae; g_ ; s_	0.10%	~~~
	o_Bacteroidales; f_; g_ ; s_	0.07%	~~~
	Bifidobacterium adolescentis	0.18%	Linked to gut-brain axis interactions
	Pseudomonas stutzeri	0.06%	Found in many different environments

## RESULTS

- A high frequency of bacterial taxa are of unknown origin, identified as k Bacteria.
- Greengenes 99% OTUs (V4 region bound by the 515F/806R primer pair) classifier.
- NCBI BLAST: Uncultured Prokaryotic Clone (83.54% Identity) Mycoplasma sp. (81.48% Identity)
- There is high individual variation between all four lab-reared spiders, some microbes more abundant than others but similarly present amongst all samples.
- Microbiome *diversity* differs between the F2 generation widows genetically originated from two separate sites.
- Microbial taxa identified as beneficial gut bacteria in humans are also present in widows.

## **FUTURE DIRECTIONS**

- We expect differences between urban, desert, and controlled lab habitats to produce microbiome differences in the next dataset.
- Goal: Identify the mechanisms that shape traits favoring urban winners.
- Looking at urban spiders may reinforce the view that cities are complex, spatially dynamic ecosystems, especially as urban widows have extensive nutrient variation [1]
- Internal microbiome comparison attempts to identify the role diet and environment play, like previous studies with covotes [2].
- Future studies will ask how spider behavior (e.g. cannibalism) shapes the microbiome. which may then shape future behavior.
- More generally, we hope to disentangle the functional importance of microbes in living organisms (e.g., metabolizing vitamins essential for reproductive success [3]).







