



Characterizing the Bacterial Gut Microbiota of Herbivorous Insects

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Introduction

- Herbivorous insects have adapted ability to process plant toxins possibly due to their gut bacteria.
- The research aims to identify and culture gut-associated bacteria from grasshoppers, beetles, and caterpillars
- Many of the gut associated bacteria are environmentally acquired, and thus more likely to be culturable than mammalian gut bacteria.

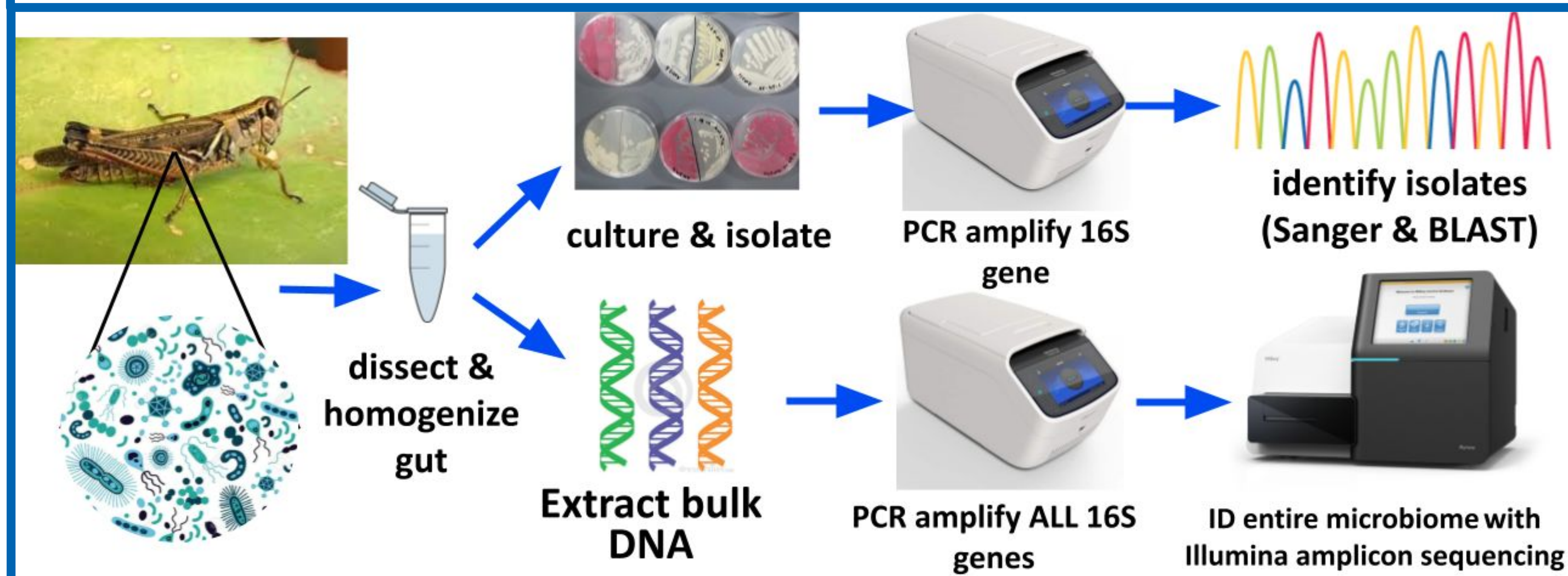
Research question: How much of the herbivorous insect microbiome can we isolate in culture?

Hypothesis: I predict that we will be able to isolate most of the bacteria genera present in insect guts using standard aerobic culture methods.

Methods

- 16S Illumina Sequencing performed on dissected & homogenized insect guts
- Culture and isolation of bacteria
- Sanger Sequencing of Bacterial Isolates
- NCBI Blasting

Illumina and Sanger Sequencing



Colorado Potato Beetle on Eggplant
Courtesy of Wikipedia

Insect Order	# of Frequent Bacterial Genera	# of Frequent Bacterial Genera with Isolates	% of Amplicon Sequencing Reads that Assign to Isolated Frequent Bacterial Genera
Beetles	19	11	54.1%
Caterpillars	24	11	40.0%
Grasshoppers	6	4	79.9%
All insects	33	19	55.2%

Table 1. Results from both Illumina and Sanger sequencing, with Frequent bacteria being genera found in greater than 20% of insect samples in each characterized order.

Results

- Bacteria from *Weissella*, *Pantoea*, *Enterobacter*, and *Pseudomonas* were isolated and cultured from insect guts.
- Comparison of bacterial isolates and illumina data reveals successful culture of known abundant genera in aerobic conditions.
- Cultured percentages: 54.1% in beetles, 40.0% in caterpillars, and 79.9% in grasshoppers, totaling 55.2% of identified bacterial genera in insect guts.

Discussion

- Abundant gut bacteria in insects may serve as primary targets for toxin degradation due to their high prevalence.
- Isolate data suggests a sizable portion of gut bacteria can be cultured aerobically, offering potential toxin degraders for further investigation.
- Certain prevalent genera like *Spiroplasma*, *Wolbachia*, and *Rickettsia* may be difficult to culture due to their intracellular lifestyles, posing challenges for toxin degradation studies.

Future Research

- In vitro cultured bacterial isolates are compared with illumina data to select potential toxin degraders.
- Selected isolates will undergo in vitro toxin degradation assays in future research.
- Specialized and Alternative culturing strategies fo uncultured isolates

References

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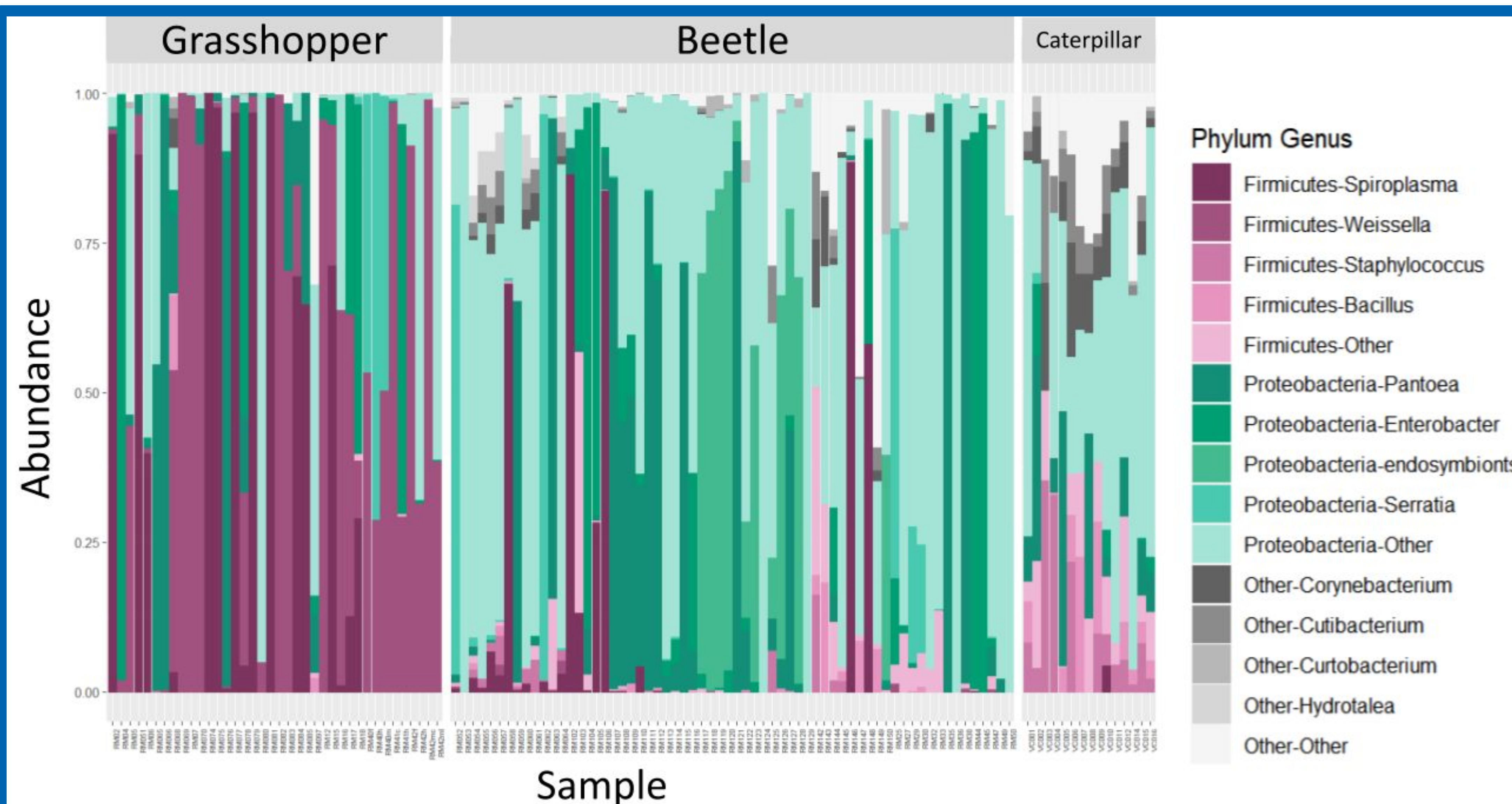


Fig 1. Bacterial relative abundance of bacterial genus compositions samples from beetles, grasshoppers, and caterpillar insect guts.