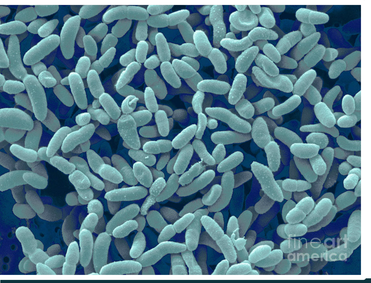

Analyzing the Genetics of Uric Acid Degradation by the Bacterium *Acetobacter fabarum*

Hiruni Dodangoda & Peter Newell
SUNY Oswego Dept. of Biological Sciences

Introduction

- The gut microbiota within organisms can not only have a great impact on the metabolism of the organism but can also influence a wide range of traits.
- *Acetobacter fabarum* is a gut bacterium that resides in *Drosophila* (1). Previous research suggests it can degrade uric acid (UA) (2), a major excretory waste product of its host.
- This study utilized genetic manipulation to test the function of predicted uricase and oxidoreductase genes in UA degradation.
- We also tested the ability of several other *Acetobacter* species to degrade UA in an agar plate assay.

Hypotheses

- Mutants with an inactive uricase gene (UriKO) will not be able to degrade UA, while mutants with an inactive oxidoreductase gene (OxiKO) may or may not, as this gene is uncharacterized.
- *Acetobacter* species isolated from *Drosophila* will possess uricase activity, leading to dissolution of UA crystals in UA agar plates.

Experiments

Genetic Manipulations:

- OxiKO and UriKO strains needed to be cured of antibiotic resistance plasmids prior to testing. First, a MIC experiment was conducted to identify the appropriate antibiotic concentration. Second, a cycolserine enrichment was performed to isolate bacteria that lack tetracycline resistance.
- The mutant strains were then grown in plates containing Uric Acid along with the wildtype as a control (data not shown).

Testing additional strains for uricase activity:

- Due to low uricase activity in the *Acetobacter* strain we were studying, we tested additional isolates using a UA plate assay in which dissolution of UA crystals indicates activity (Fig. 1).

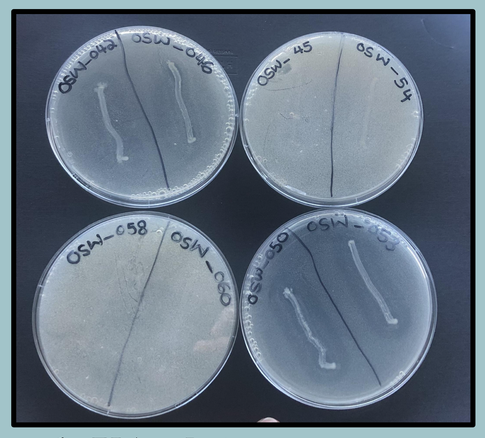

Figure 1. UA plate assay.

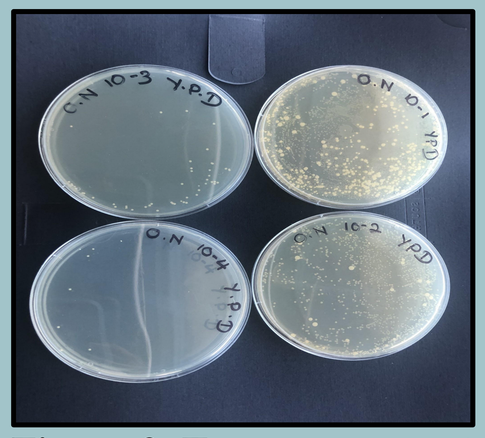

Figure 2. Transposon mutants

Alternative approach: Isolate mutants of *Acetobacter #50*

- The plate assay showed low uricase activity of wildtype *A. fabarum* (Fig. 1; OSW_54) but high activity for *Acetobacter* strain 50 (OSW_50). Therefore, we began the process of isolating uricase mutants of strain 50 using transposon mutagenesis (Fig. 2).

Results

- Genetic and phenotypic data suggest successful isolation of OxiKO and UriKO mutants. However, low uricase activity for wildtype of this strain made further study difficult.
- UA plate assay showed significant uricase activity for *Acetobacter* strains 42, 46, 50 and 53.
- Trial genetic manipulations of *Acetobacter* 50 were successful, suggesting that isolating uricase mutants in this strain will be feasible.

Conclusions

- *Acetobacter* gut bacteria isolated from *Drosophila* fruit flies show uricase activity in an agar plate assay.
- Transposon mutagenesis successfully produced mutant bacteria for strain #50.
- Future research will target the uricase gene of *Acetobacter #50* to test if it is required for UA degradation.
- UA is a major waste product in *Drosophila* as well as humans (3). Future experiments will test the impact of microbial uricase activity on *Drosophila* health.

References & Acknowledgements:

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