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Exploring the Growth Rates of Different Bacterial Isolates Found in Purple Pitcher Plants (*Sarracenia purpurea*)

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Abstract

Microbial communities are an essential part of every ecosystem, with bacteria within these microbial communities playing a crucial role in nutrient cycling. But research is lacking concerning how different microbial taxa vary in growth rate and enzyme production. By beginning to understand these microbial processes we can further develop our understanding of nutrient cycling within ecosystems, and how they promote environmental stability. We turned to *Sarracenia purpurea* (purple pitcher plant) as a model system for community ecology. The microbial communities within the plants can grow in a naturally enclosed and initially sterile environment, where then their growth can be monitored. By looking at the trends in microbial growth rates in a controlled laboratory environment, we can improve our knowledge of what tradeoffs might exist and how microbes may influence pitcher plant success. Our study asked: Is there a relationship between microbial growth rate and taxonomy or enzyme activity?

To explore this question, we looked at 34 bacterial strains collected from purple pitcher plants with known taxonomy. By growing them from frozen cultures in R2A media we measured the bacterial density of each strain (in triplicate) over 48 hrs at 590 nm. We compiled the growth curve data with our previously collected enzyme and taxonomy data and processed our analysis in R. From this analysis, we concluded that there were no significant differences in growth rates between different genera, families, or orders when the 34 bacterial isolates were clustered by taxonomy. There were also no significant differences in mean growth rate with protease, chitotriosidase, and endochitinase activity. But we did see some weak net negative directional effects in the probability densities for lipase and β -N-acetylglucosaminidase activity based on our model, which may indicate a potential tradeoff with the growth rate for the bacteria.

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Introduction

Microbial communities are an essential part of every ecosystem [1]. The bacteria within these microbial communities play a crucial role in nutrient cycling within their systems. In host-associated systems, bacteria may produce enzymes that the host system cannot [2]. Bacteria may face tradeoffs between growth and enzyme production, as both are costly.

To better understand the relationship between growth rates, taxonomy, and enzyme production, we turned to *Sarracenia purpurea* (purple pitcher plant), a model system for community ecology [3]. These plants rely heavily on prey capture and aquatic microorganisms to access essential nutrients [4]. Research is lacking with regard to how different taxa of microbes vary in growth rate and enzyme production. This information may be critical for understanding nutrient cycling within the pitcher plant system. By looking at the trends in microbial growth rates in a controlled, laboratory environment, we can improve our knowledge of how microbes may influence pitcher plant success.

Question/Hypothesis

We asked: **Is there a relationship between microbial growth rate and taxonomy or enzyme activity?** We hypothesized that there would be a difference in the average growth rate based on bacterial Order.

Materials and Methods

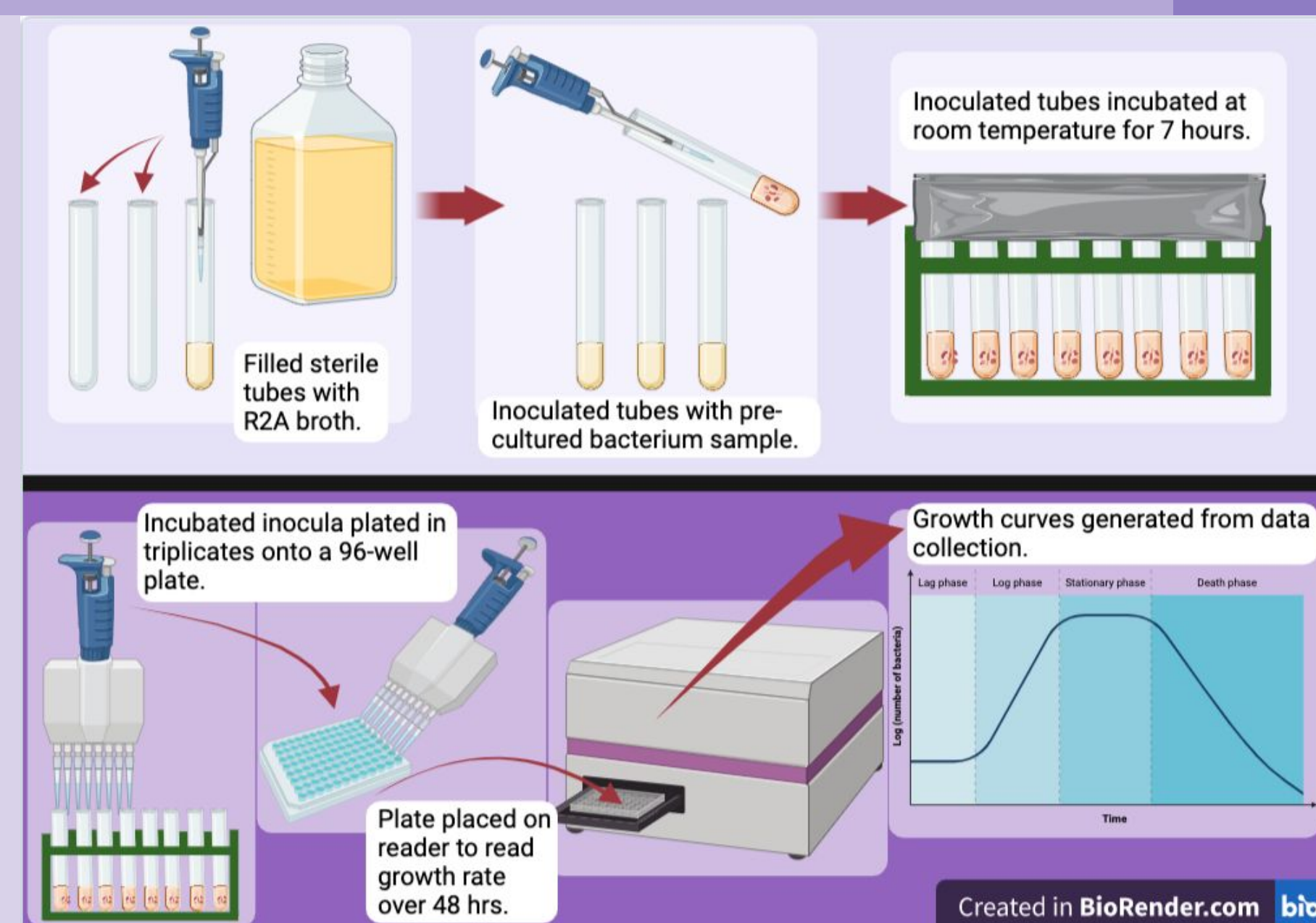


Figure 1. A diagram showing the process of collecting optical density measurements on bacterial isolates.

34 unique bacterial isolates from pitcher plant communities were:

1. Grown in R2A media from frozen culture
2. Measured optical density over 48 hrs at 590 nm
3. Optical density data was collected in triplicate
4. Growth curves were analyzed in R using GrowthCurveR package
5. Rates were compared across taxonomic groups and with previously collected enzyme data

Results

No significant difference in growth rate based on taxonomy

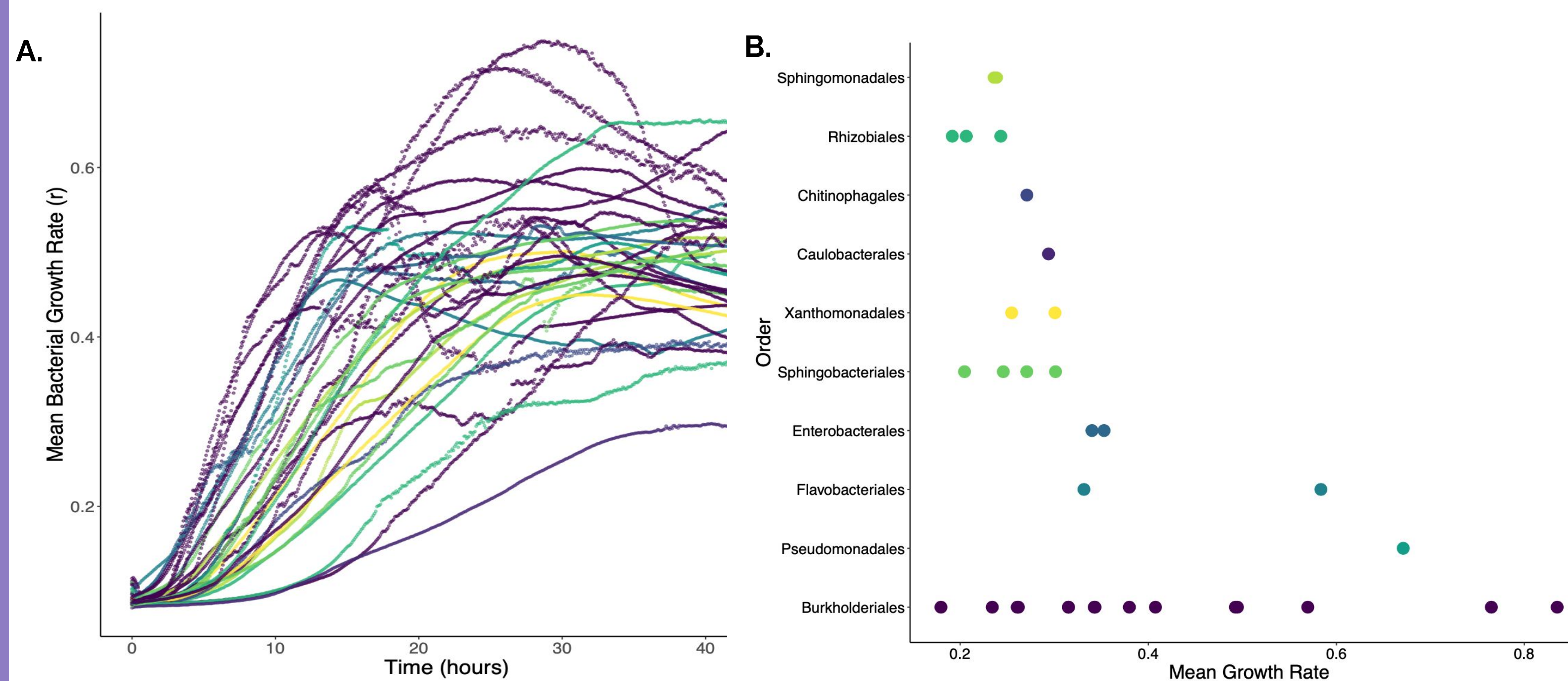


Figure 2. Comparing the growth curves and growth rate for 34 pitcher plant bacterial isolates using optical density (590 nm). A) Bacterial optical density measurements taken over 48-hours. We saw no significant differences between triplicates, so each point is mean absorbance, colored by order. B) Logistic regressions were fit to the OD data and growth rate for each isolate was calculated using the GrowthCurveR package in R. We found no significant differences in mean growth rate (r) based on taxonomy of the bacterial isolate (genus, family, order).

Weak directional effects of enzyme activity on growth rate

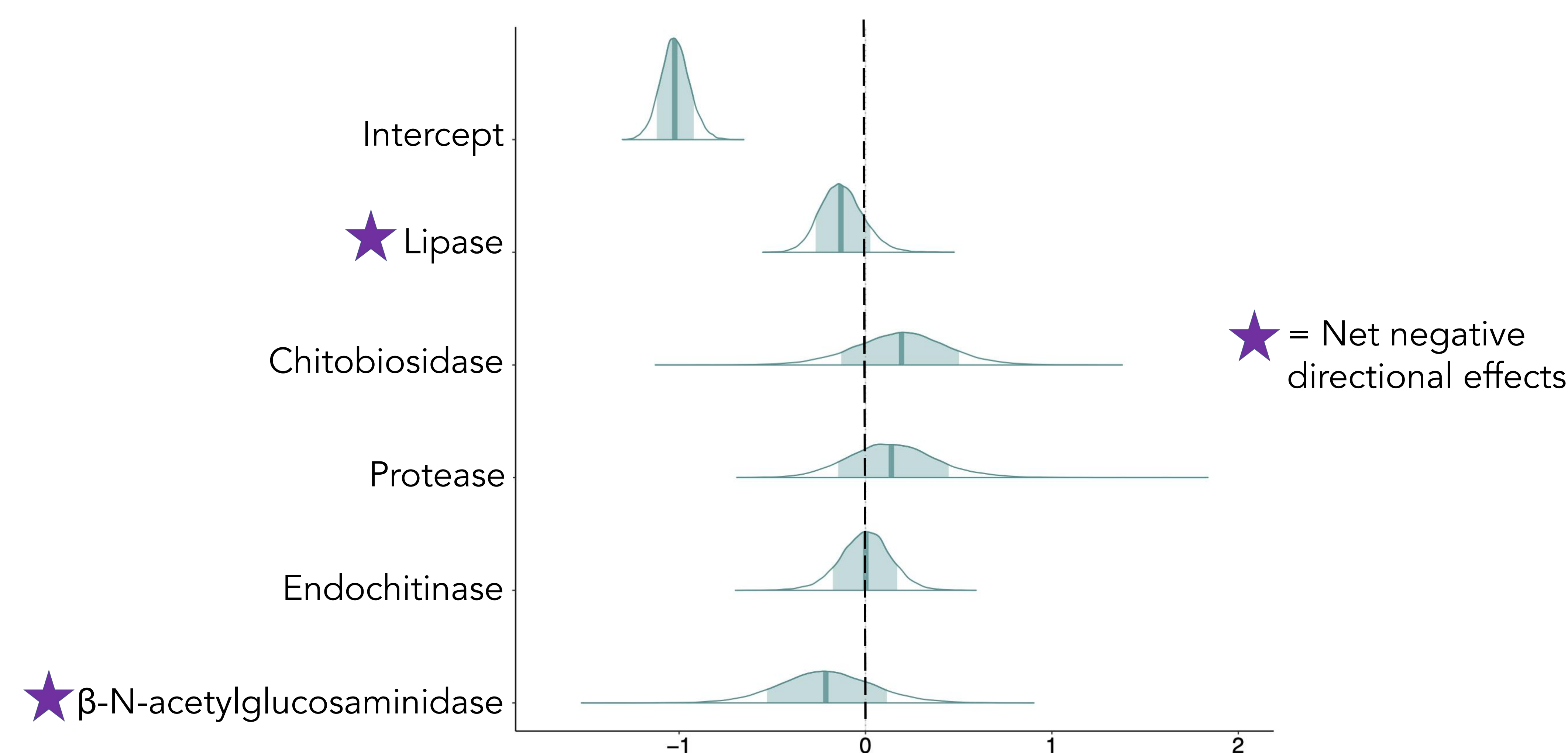


Figure 3. Hydrolytic enzyme activity (scaled and centered) may be impacted by growth rate for pitcher plant bacterial isolates. Each distribution is the probability density of a specific enzyme's effect while other predictor variables are held at their means. Lipase and β -N-acetylglucosaminidase activity show a net negative directional effect (86% and 80%, respectively) on bacterial growth rate when compared to the mean intercept (dashed line at zero).

Conclusions

- ❖ Pitcher plant bacterial isolates grew well in R2A media.
- ❖ Using microwell plates to collect data on bacterial growth curves had reproducible results.
- ❖ There were no significant differences in growth rates between different genera, families, or orders when the 34 bacterial isolates were clustered by taxonomy.
- ❖ We saw no significant differences in mean growth rate with protease, chitobiosidase, and endochitinase activity.
- ❖ We did see some weak net negative directional effects in the probability densities for lipase and β -N-acetylglucosaminidase activity based on our model.
 - Indicating a potential tradeoff with growth rate for the bacteria.

Further Information

In future experiments we plan to increase the number of samples for each taxonomic group in our study. It's difficult to establish significant or non-significant differences in growth rates and enzyme activity with only 34 samples and some taxonomic groups with only one member. A phylogenetic analysis would complement this approach.

However, it would also be interesting to pursue a study on how bacterial growth rate is influenced by other conditions, such as an increase in temperature. Climate change effects may influence bacterial growth and activity in their ecosystems.

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