Measuring Growth Quality of Bacteria Using Phenotypic Microarrays

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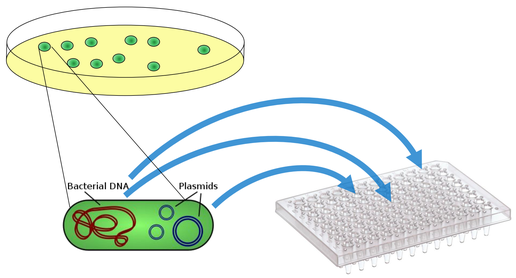
Introduction

Phenotypic microarrays (PMs) provide a large-scale, high-throughput method for measuring growth of microorganisms over time.



Growth curves are produced from the OD600 absorbance measurements recorded by the PM instrument for each well.

Each well consists of minimal media and a unique carbon- or nitrogen-based substrate.

Over 96 wells, various phenotypes can be captured with respect to several growth conditions.

Goals

Develop an automated analysis pipeline to accurately qualify growth of bacterial cells using PM measurements.

Using logistic growth curve parameters, quantify the growth level of various bacterial cells.

Apply this analysis toward biological discovery and phenome defintions.

Computational Methods

QC Preprocessing

Growth curves from PM undergo filtering to remove troublesome data from analysis pipeline:

$$Filter\ 1:\ OD_{30minutes} \geq 0.25$$

$$Filter\ 2:\ OD_{1hour} \geq OD_{24hours} - 0.03$$

Data captured by these filters should be closely examined for experimental error.

Logistic Model

Growth curves fitted by a logistic model:

$$Y = y_{min} + \frac{A - y_{min}}{1 + e^{\left(\frac{\mu}{A}(\lambda - t) + 2\right)}}$$

y_{min}	*background*
A	*asymptote*
μ	*max. growth rate*
λ	*lag phase*
t	*time*

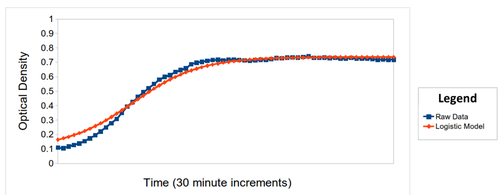


Figure 1. Raw growth curve (blue) fitted with a logistic model curve (red).

Harmonic Mean

To quantify and represent growth quality, the *harmonic mean* is calculated on the logistic model:

$$HM = \frac{N}{\sum_{i=1}^{N} \frac{1}{y_i + A}}$$

Results

Metabolic Assessment

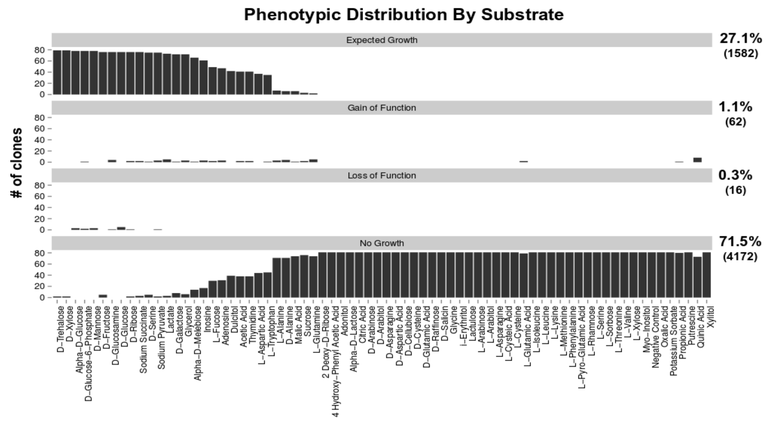


Figure 2. Phenotype distribution of 72 unique carbon growth conditions across 81 *E. coli* clones, each inoculated with a unique unknown putative bacteriophage gene. Growth assessment can give insight into possible metabolic effects from the genes under investigation.

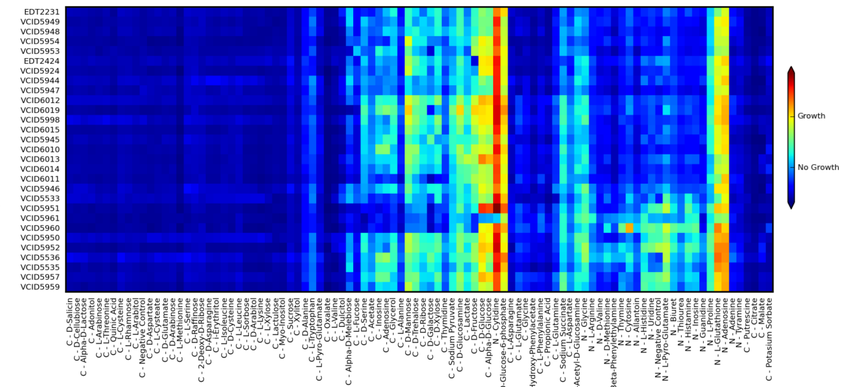


Figure 3. Qualitative growth level evaluation across a mix of 96 carbon- and nitrogen-based substrates using harmonic mean calculations.

Future Directions

Optimize QC and model prediction analysis pipeline.

Implement pipeline into the Viral Dark Matter website for real-time automated analysis and data visualization.

Provide reconciliation support for *in silico* metabolic networks encompassing a wide variety of microbial species when using flux balance analysis systems.

Acknowledgements: Viral Dark Matter Consortium, Edwards Lab, Rohwer Lab, Segall Lab, SDSU Biomath Group

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