Understanding and Directing Population Dynamics and Metabolic Switching in *Escherichia coli*

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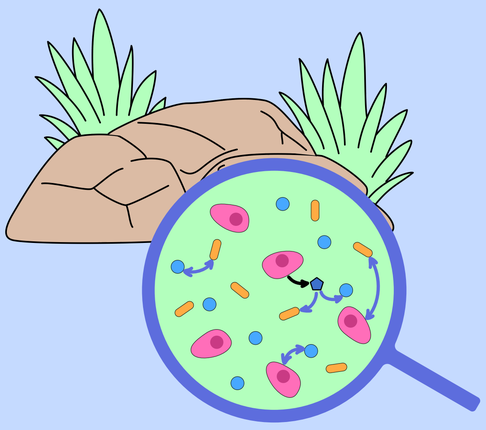
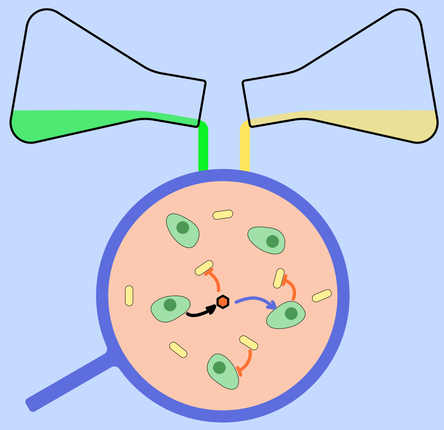
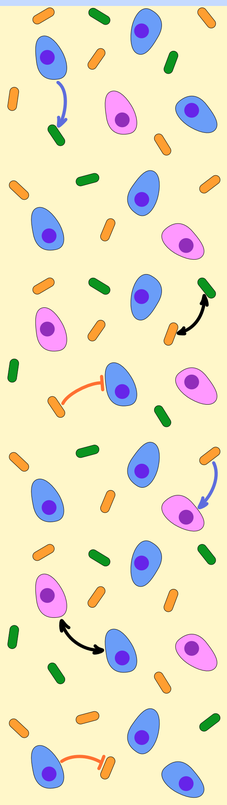


CONTEXT

Natural microbial communities tend to self-regulate, whereas synthetic microbial communities often struggle to maintain population balance over time, requiring external control strategies[1].

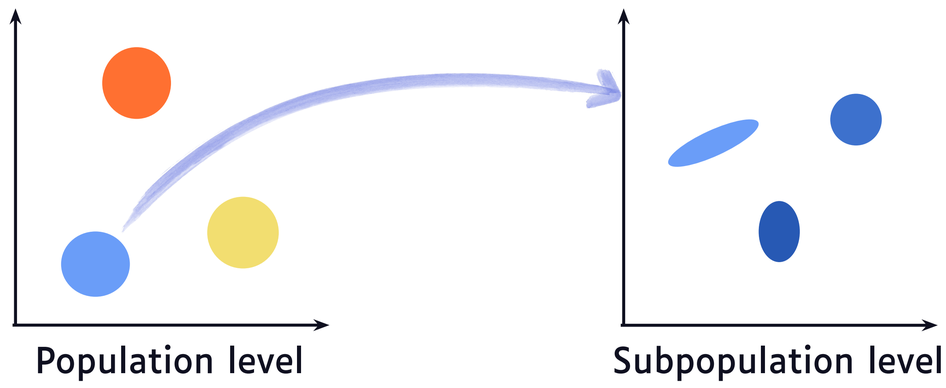




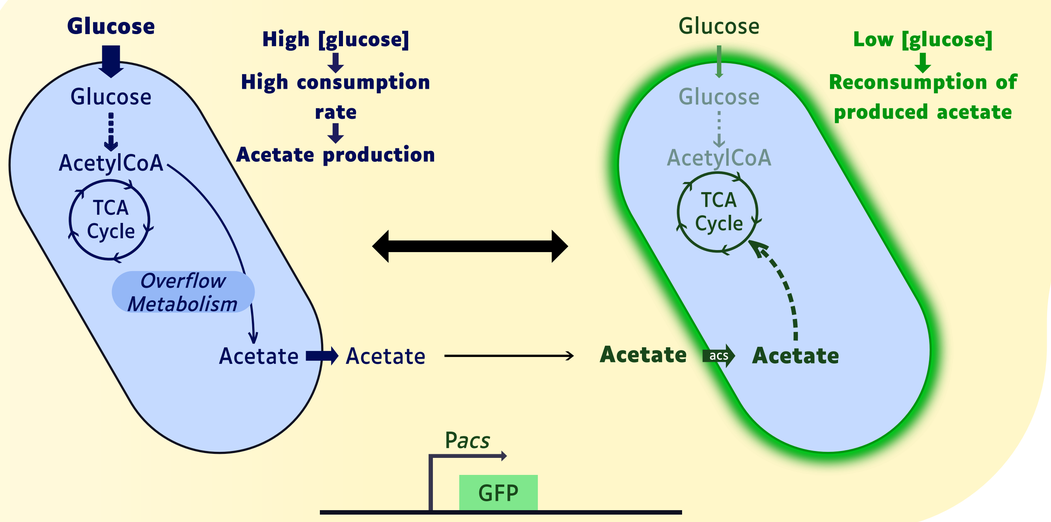
INTRA SPECIES DYNAMICS

An important yet often overlooked factor in microbial community stability is the presence of phenotypic diversification (i.e., subpopulations). These subpopulations can arise from distinct metabolic states (e.g., carbon consumption, stress responses)[2], leading to heterogeneity within the population that can influence interspecies interactions.

Studying these subpopulations is key to stabilizing synthetic communities. We used the diauxic shift between glucose and acetate consumption in *Escherichia coli* as a model. Such metabolic state transitions are monitored using flow cytometry and GFP-based reporters.



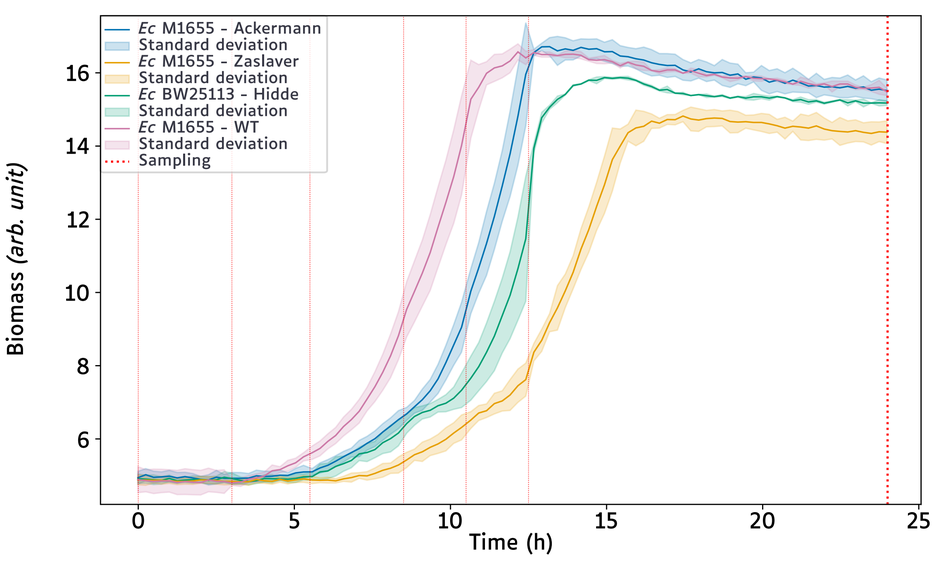
MODEL



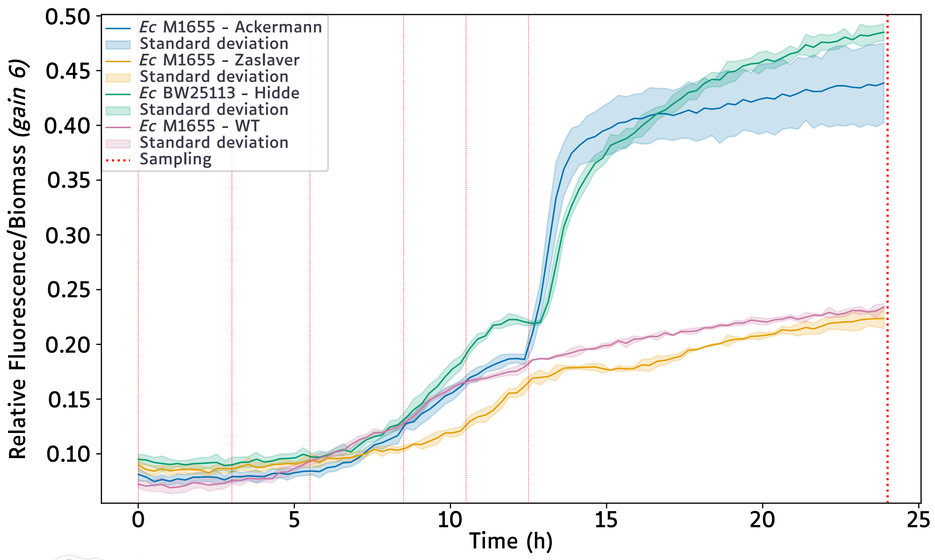
PRELIMINARY RESULTS

Characterisation of strains was performed using a BioLector (microplate-based bioreactor), enabling real-time monitoring of growth and fluorescence.

Biomass per strain over time

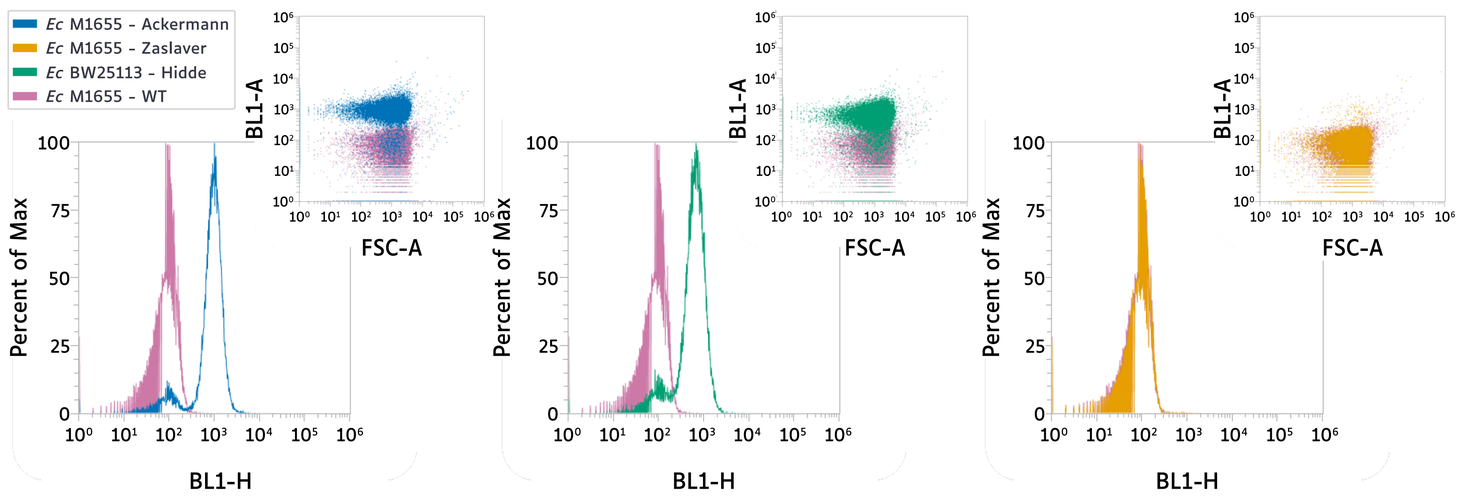


Relative Fluorescence/Biomass *(gain 6)* per strain over time



FLOW CYTOMETRY

Fluorescence (GFP) comparison of engineered and WT strains at 24h (Attune cytometry)



PERSPECTIVES

1. Evaluate additional strains and select a candidate based on carbon consumption and fluorescence signal intensity
   Characterise dose–response behaviour during the diauxic shift
2. Continuous culture in a bioreactor with the selected strain
   Online fluorescence monitoring by flow cytometry
3. Introduction of a bacteria or yeast to investigate interspecies dynamics and their impact on subpopulations

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[1] Martinez, J. A., et al, Automated Adjustment of Metabolic Niches Enables the Control of Natural and Engineered Microbial Co-Cultures. *Trends Biotechnol.* **2025**, S0167779924003652. https://doi.org/10.1016/j.tibtech.2024.12.005.

[2] Delvigne, F., et al, Metabolic Variability in Bioprocessing: Implications of Microbial Phenotypic Heterogeneity. *Trends Biotechnol.* **2014**, 32 (12), 608–616. https://doi.org/10.1016/j.tibtech.2014.10.002.