The multi-level relationship between heterotrophic bacteria and nutrients

Diauxie and co-utilization are not exclusive during growth in nutritionally complex environments

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The classic view of microbial growth strategy when multiple carbon sources are available states that they either metabolize them sequentially (diauxic growth) or simultaneously (co-utilization).

”**All you can eat**” *vs.* ”**a la cart**” strategies result in different growth dynamics (Monod, 1942).

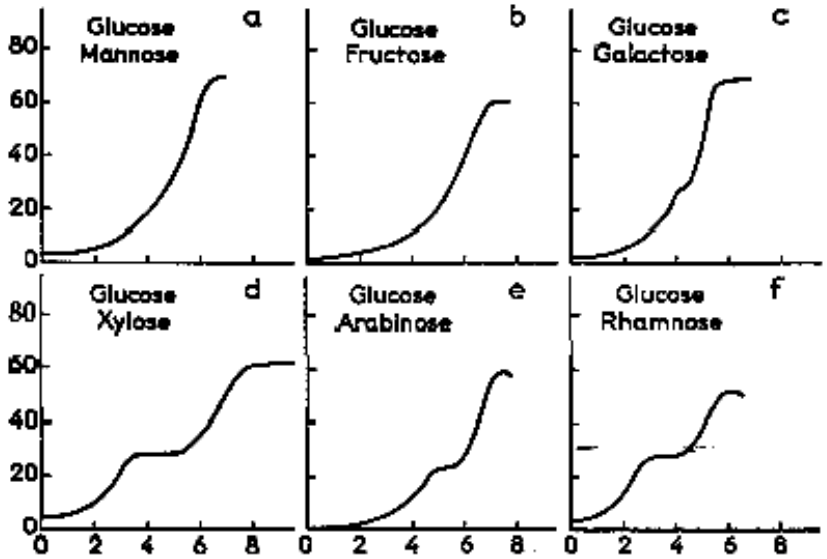
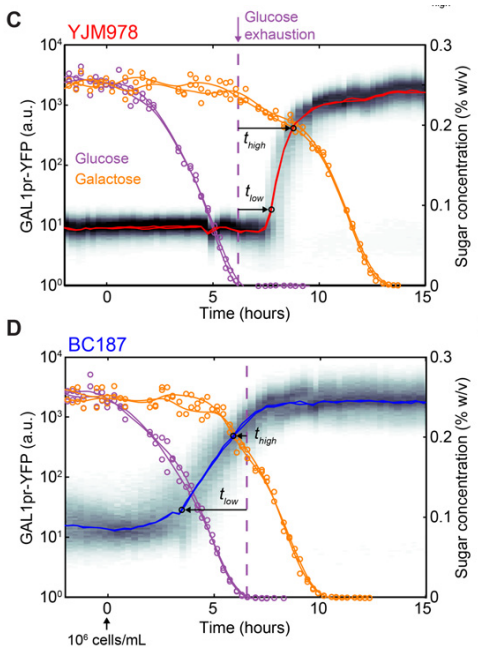
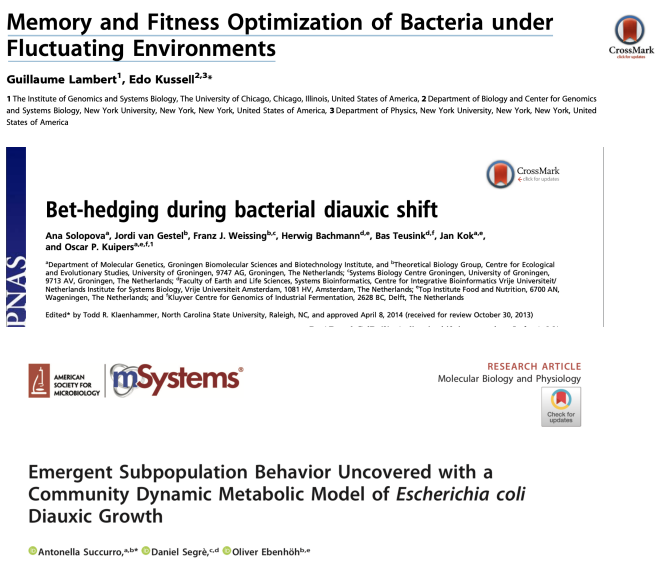


Fig.1. Growth of *Escherichia coli* in the presence of different carbohydrate pairs serving as the only source of carbon in a synthetic medium[30].

Diauxic shift is commonly seen as a phase in which the bacteria prepare themselves to use the second sugar. The existence of two stable cell types with alternative metabolic strategies emerge and coexist in a culture of the bacterium has been demonstrated.

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Memory and Fitness Optimization of Bacteria under Fluctuating Environments

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Bet-hedging during bacterial diauxic shift

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RESEARCH ARTICLE
Molecular Biology and Physiology

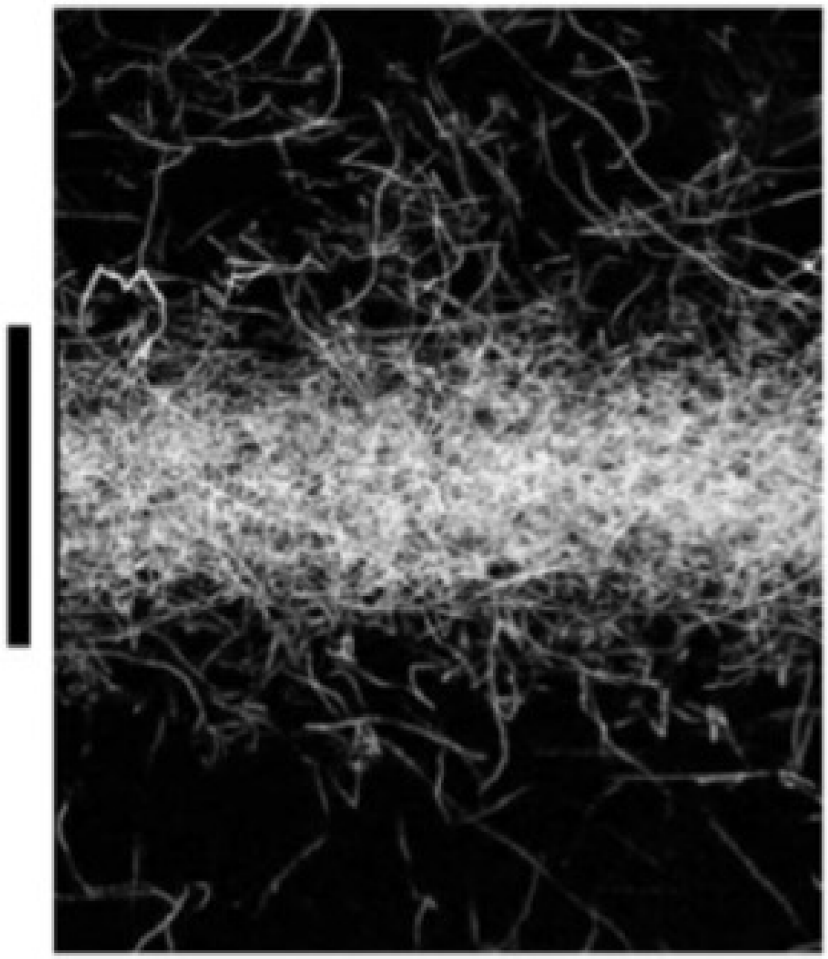
Emergent Subpopulation Behavior Uncovered with a Community Dynamic Metabolic Model of *Escherichia coli* Diauxic Growth

Antonella Succurro,[a,b]* Daniel Segrè,[c,d] Oliver Ebenhöh[b,e]

Our knowledge is biased by the fact that this process has been mainly analyzed in **over-simplified laboratory settings,** i.e. using a few model microorganisms and growth media containing only two alternative compounds.

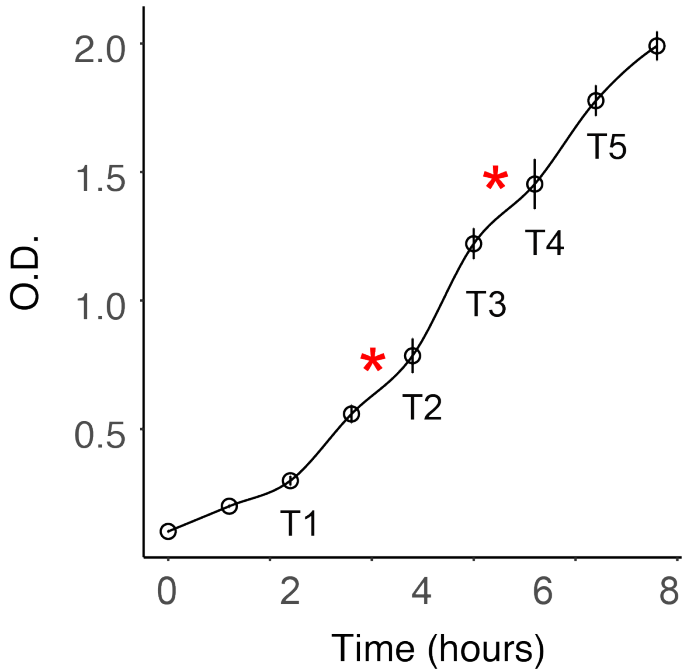
The marine environment:

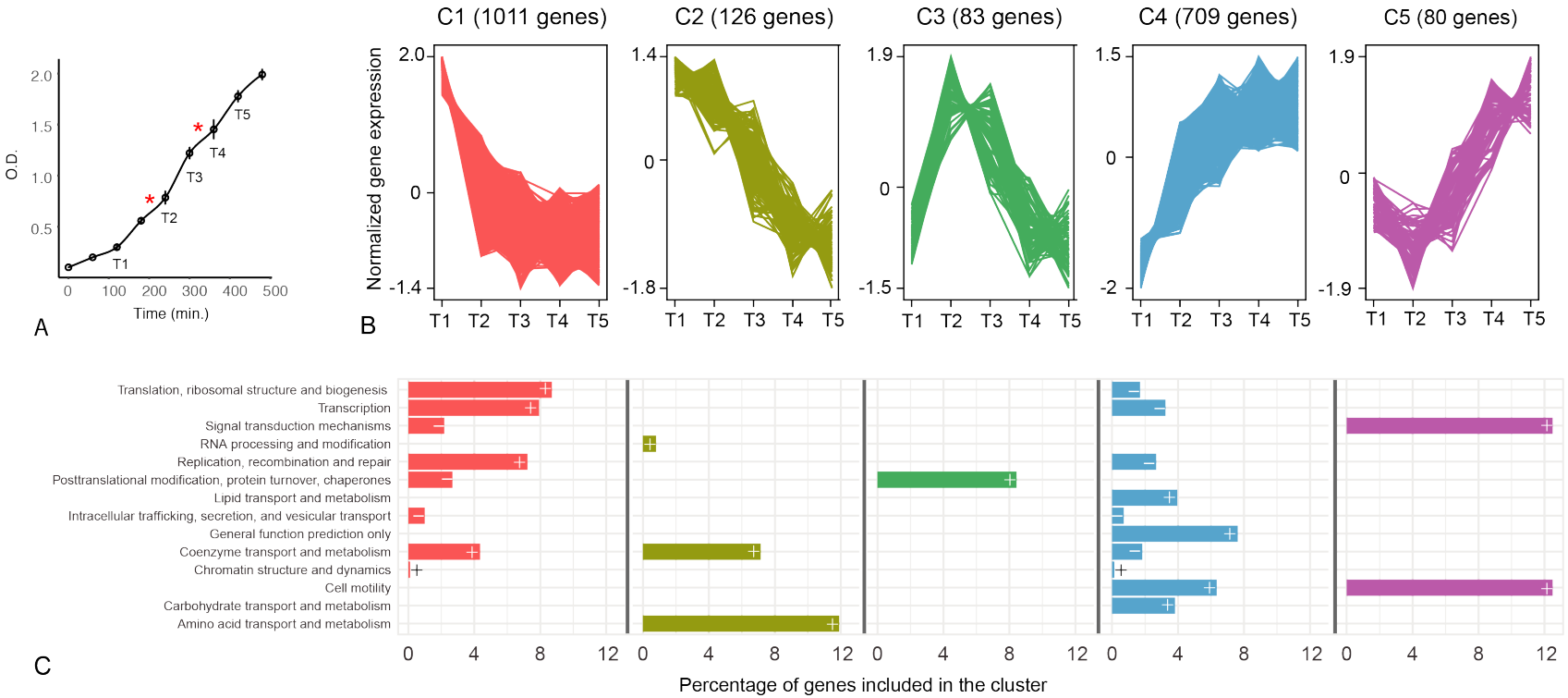
- Low average nutrient level (e.g., the concentration of amino acids is in the range of about 10^{-9} M)
- Nutrients appear and disappear in a sporadic fashion.
- DOM as a complex medium



- Chemotaxis of *Pseudoalteromonas haloplanktis* TAC125 (PhTAC125) toward a pulse of phytoplankton exudates (Stocker et al. 2010, Science)
- What happens once the patch is colonized?

We grew PhTAC125 in a complex medium (peptone, 7 replicates) and analysed its growth features.

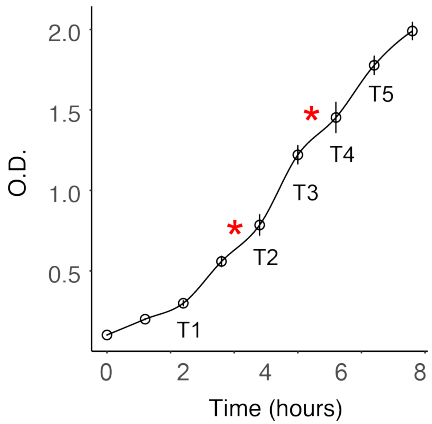


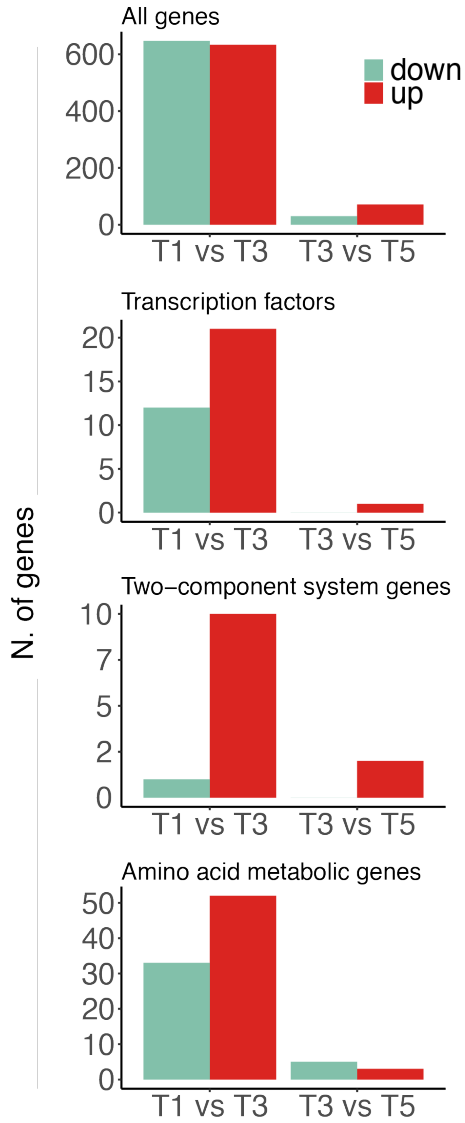


- High expression of growth-related genes in the first hours
- Stress related genes up between T1 and T2
- Motility-related genes up at the end of the growth

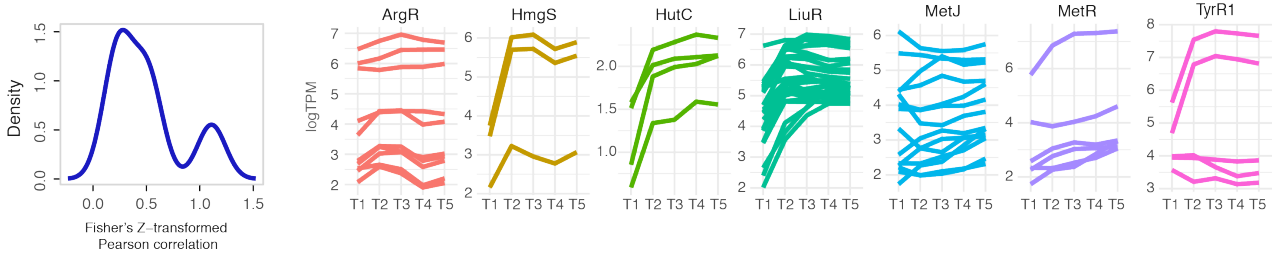
A non *E. coli*-like response to growth lag

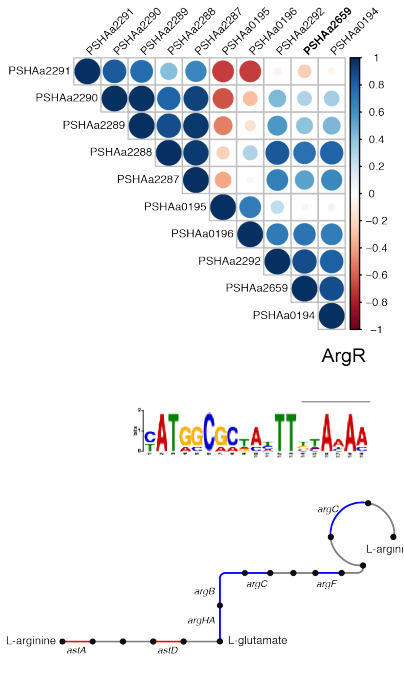
- 81 TF known to dcontrol central metabolic enzymes in *E. coli*
- Homologs for 34 of them in PhTAC125, 8 of them differentially expressed (10%)
- Only RpoS and RpoD are differentially expressed among 8 selected *E. coli* global regulators

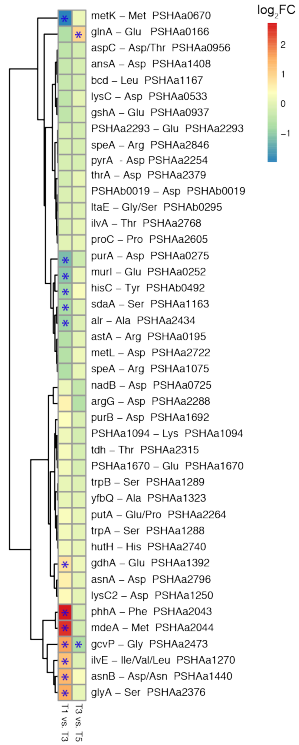




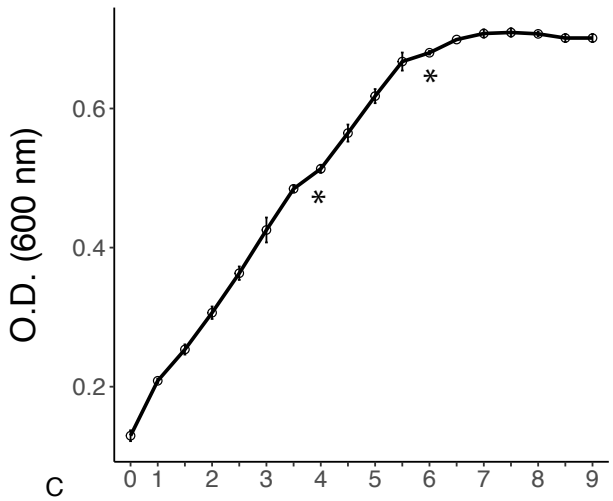
A focus on amino acids degradation. PhTAC125 is know to thrive preferentially on amino acids (Wilmes et al. 2010).







We assembled a defined, nutritionally rich medium including 19 amino acids ([0.2 mM], Cys not included) and analysed growth and metabolic features.



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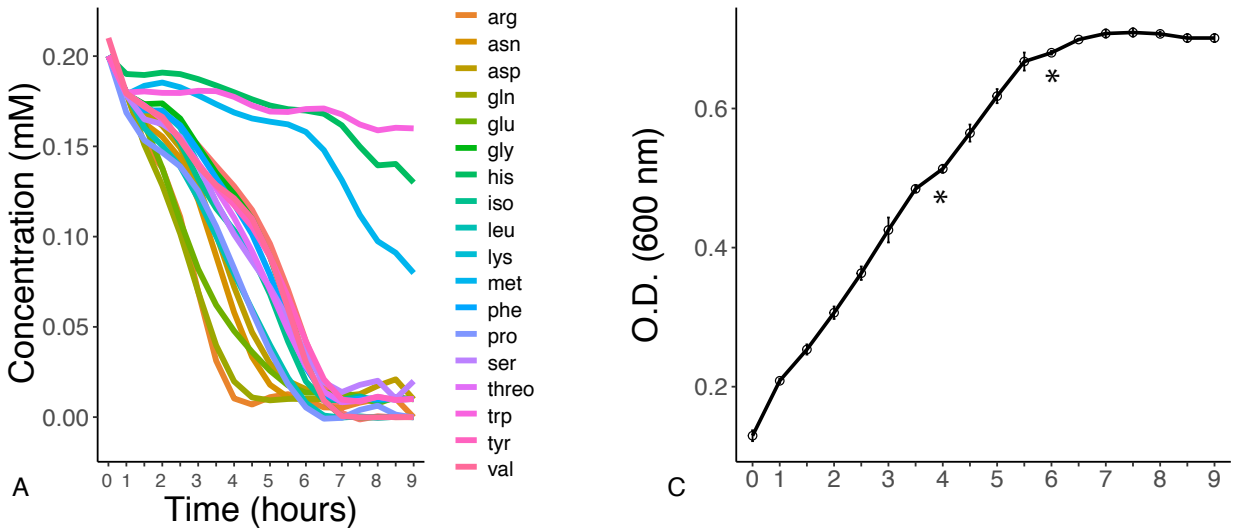

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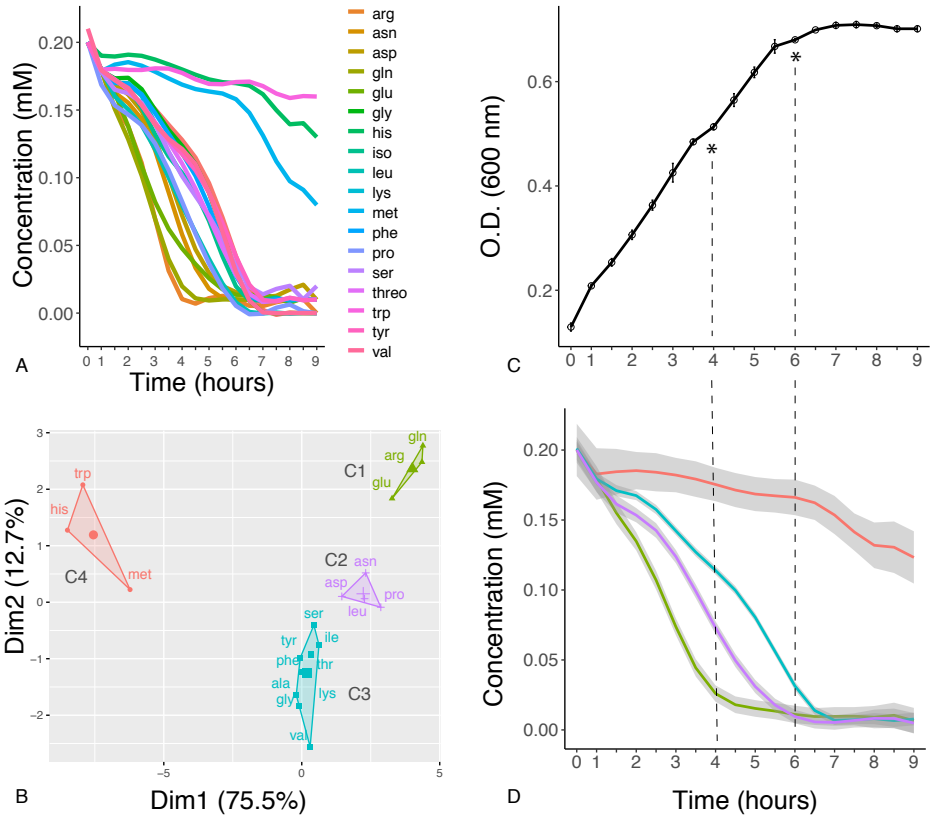

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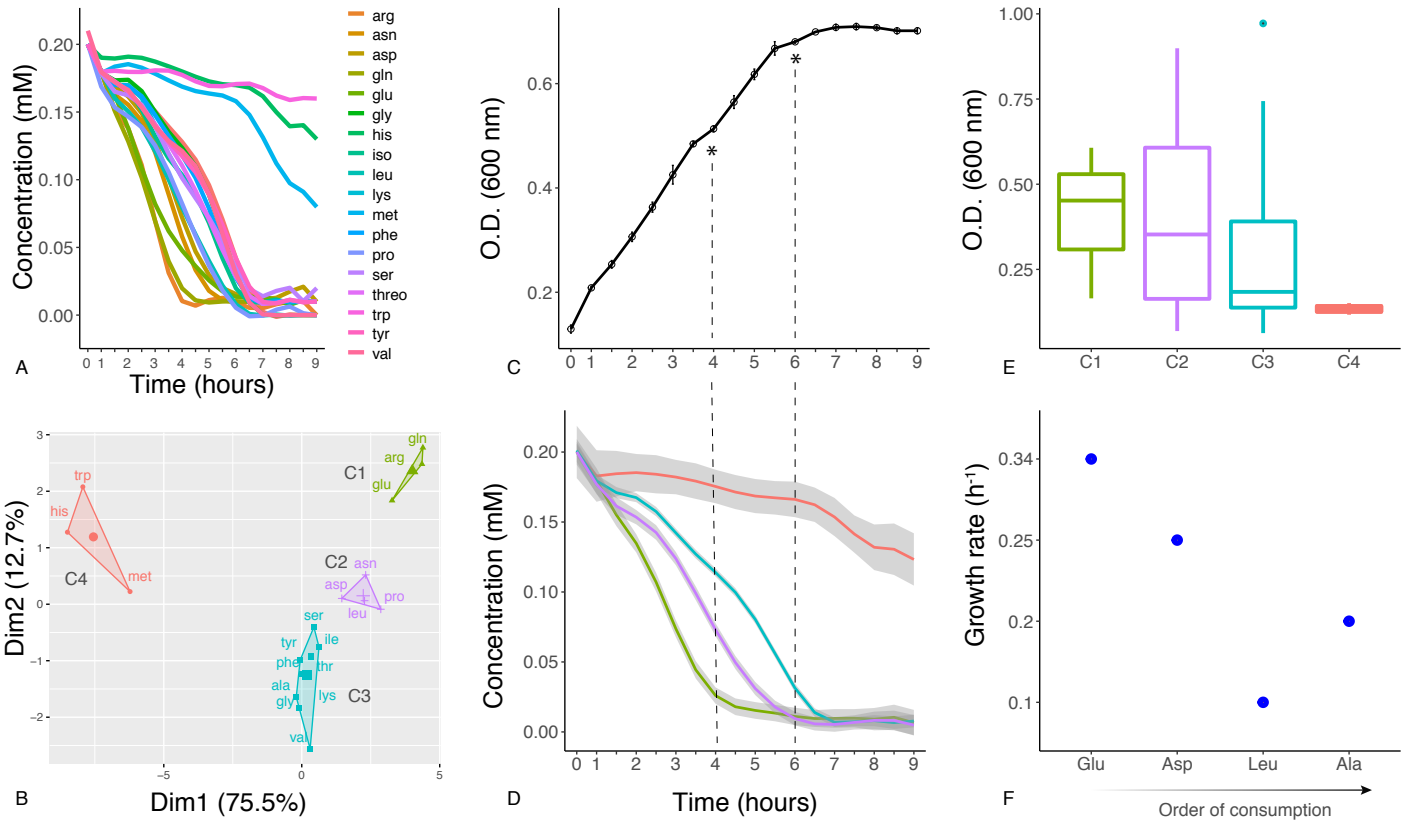


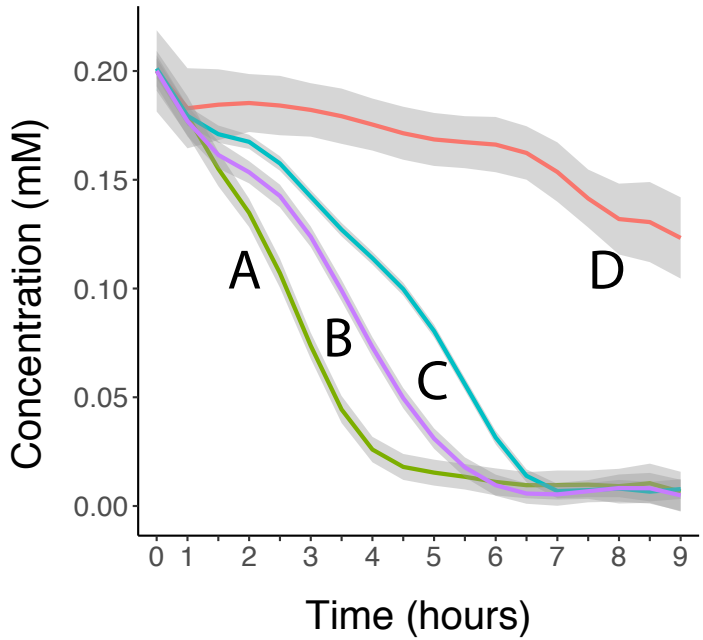
What is regulating the system?

- Uptake kinetics (no regulation, dynamics explained by different uptake kinetics).
- Active regulation The uptake of the different groups is *somehow* regulated.

What is regulating the system?

- **Uptake kinetics** (no regulation, dynamics explained by different uptake kinetics).

$$P + A \xrightarrow{\frac{\beta_A \cdot \phi_A}{k_A + \phi_A}} 2P$$
$$P + B \xrightarrow{\frac{\beta_B \cdot \phi_B}{k_B + \phi_B}} 2P$$
$$P + C \xrightarrow{\frac{\beta_C \cdot \phi_C}{k_C + \phi_C}} 2P$$
$$P + D \xrightarrow{\frac{\beta_D \cdot \phi_D}{k_D + \phi_D}} 2P$$



$$P + A \xrightarrow{\frac{\beta_A \cdot \phi_A}{k_A + \phi_A}} 2P$$
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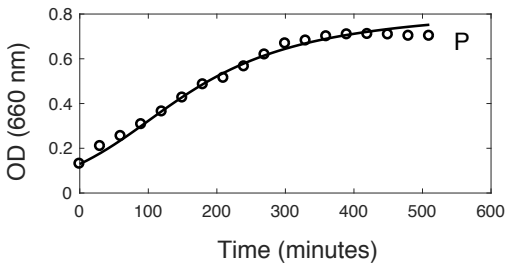
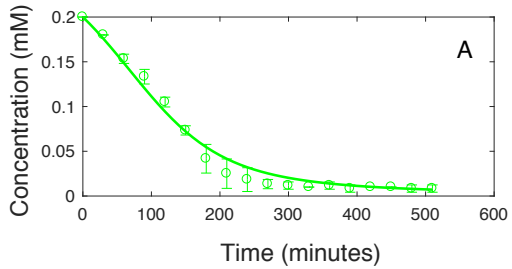
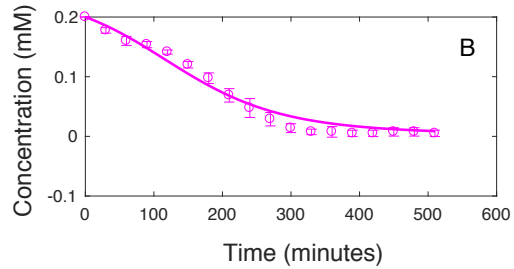
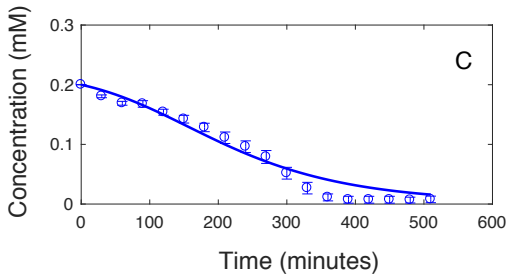
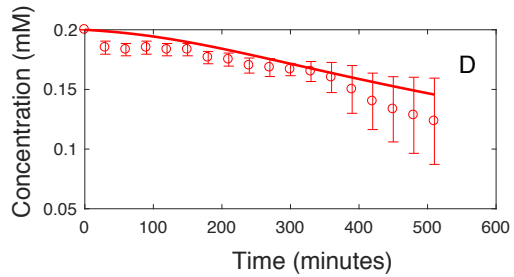
$$P + C \xrightarrow{\frac{\beta_C \cdot \phi_C}{k_C + \phi_C}} 2P$$
$$P + D \xrightarrow{\frac{\beta_D \cdot \phi_D}{k_D + \phi_D}} 2P$$











What is regulating the system?

- **Active regulation** The uptake of the different groups is *somehow* regulated. A cybernetic model (Kompala et al. 1994) is implemented.

$$P + A \xrightarrow{E_1,\ \rho_1} 2P$$
$$P + B \xrightarrow{E_2,\ \rho_2} 2P$$
$$P + C \xrightarrow{E_3,\ \rho_3} 2P$$
$$P + C \xrightarrow{E_4,\ \rho_4} 2P$$

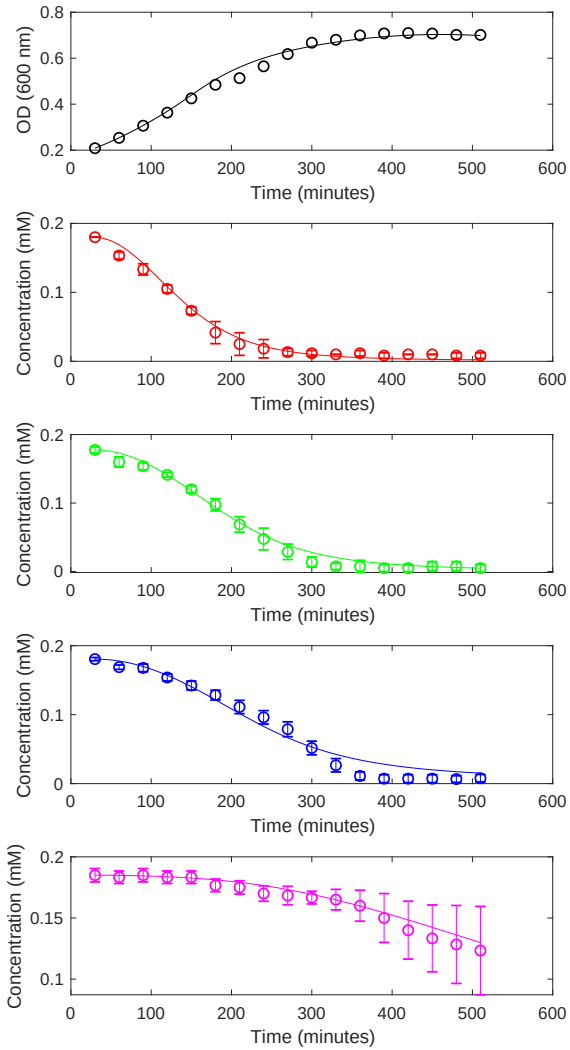
$$C \xrightarrow{A,\ \lambda_1} C + E_1$$
$$C \xrightarrow{B,\ \lambda_2} C + E_2$$
$$C \xrightarrow{C,\ \lambda_3} C + E_3$$
$$C \xrightarrow{D,\ \lambda_4} C + E_4$$

$$\frac{d\phi_{E_i}}{dt} = \lambda_i * u_1 - (\rho_1 * v_1 + \rho_2 * v_2 + \rho_3 * v_3 + \rho_4 * v_4) * \phi_{E_i} - \beta\phi_{E_i}$$

$$\frac{d\phi_P}{dt} = (\rho_1 * v_1 + \rho_2 * v_2 + \rho_3 * v_3 + \rho_4 * v_4) * \phi_P - \alpha\phi_P$$

$$\frac{d\phi_{S_i}}{dt} = \rho_i * v_i * \phi_P$$

With i = 1,2,3,4 and S = {A, B, C, D}



	MM	Cybernetic
Biomass	0.997	0.994
Cluster A	0.9893	0.9983
Cluster B	0.9967	0.9977
Cluster C	0.9947	0.9904
Cluster D	0.9723	0.9908

R^2 calculation of the fit

- PhTAC125 growth on complex media results in a triauxic growth with a regulation response to nutrient starvation that does not resemble the model one.
- Growth lag phases are due to nutrient switching.
- Sequential and co-utilization are not exclusive. Most efficient C sources are consumed first.
- A simple mathematical model based on Michaelis-Menten kinetics uptake does not result in a perfect fit with the data. Regulatory mechanisms must be included to explain this growth dynamic at the single cell level
- Emergent sub-populations?

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