

Dynamic Control of Microbial Co-Cultures

Automated Niche Adjustment for Continuous and Fed-Batch Bioprocessing

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1 MICROBIAL INTERACTIONS and AAMN

GOAL Currently, the use of modified strains are spread for synthetic biopathways and production of specific metabolites. However, it can lead to metabolic burden on modified cells [1]. An alternative is setting a labor division strategy by the use of microbial consortia to decrease metabolic burden [1].

STRATEGY

The control of consortia in co-cultivation using **Automated Adjustment of Metabolic Niches** (AAMN) is achieved in our lab with the **Segregostat** system [2].

- A** Bioreactor with co-culture (2 species)
- B** Segregostat with online flow cytometry monitoring phenotypic
- C** Results showing co-culture strains and applied regulation rules
- D** Cultivation adjustment via metabolic niches pulsing (carbon source) to regulate strains proportion

RESULTS

CASE 1 : Competition

Escherichiae coli & *Saccharomyces cerevisiae*

CASE 2 : Commensalism

Kazachstania bulderi & *Lactobacillus plantarum*

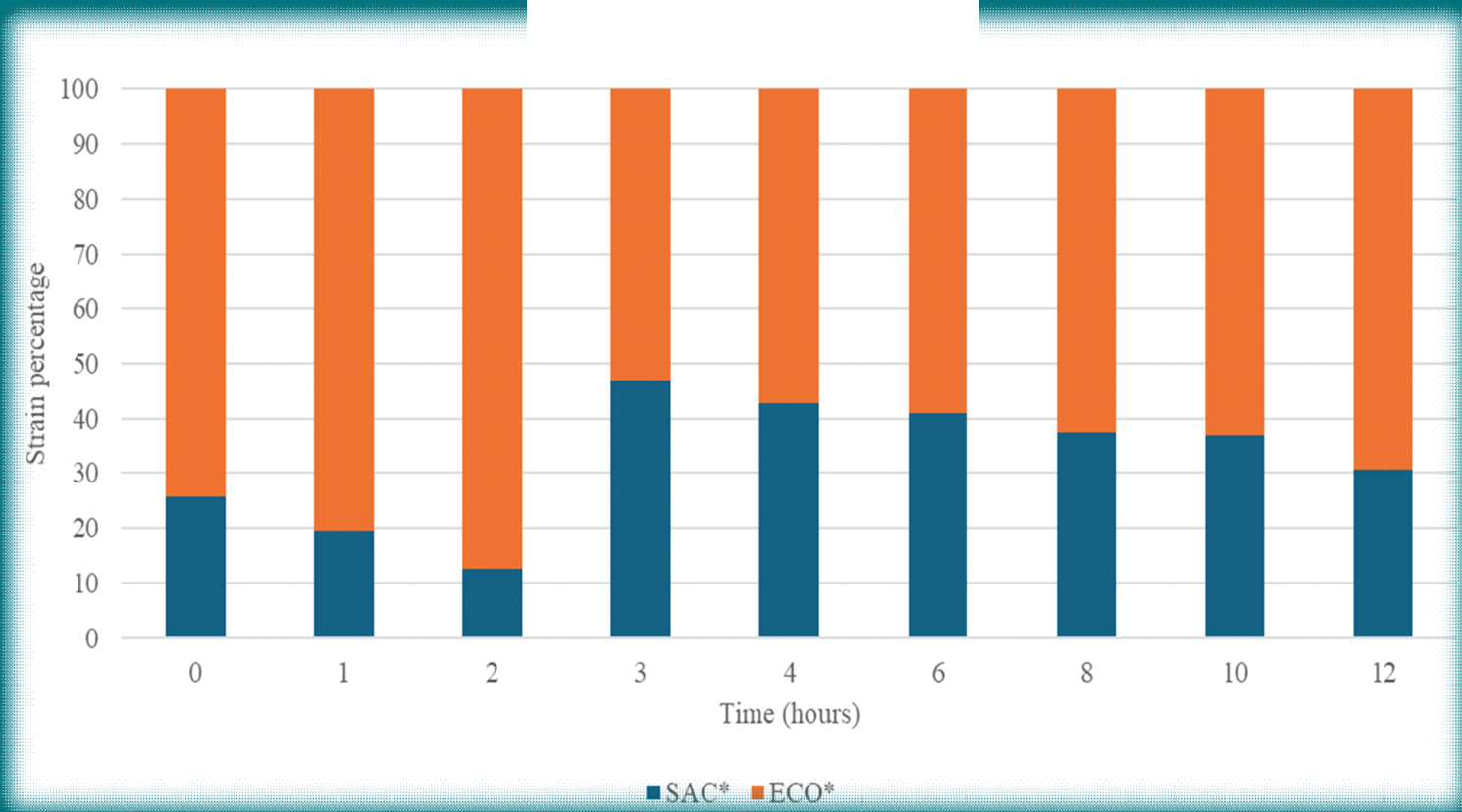
CASE 3 : Proof of concept

Production yield increased by 3.5-fold with AAMN control [2]

Elementary Modes, derived from genomes, design metabolic niches, compute competitiveness, and simulate co-culture using Monod-type (MONCKs)[2]

2 ADAPTATION TO FED-BATCH

The **AAMN** strategy was adapted to Fed-Batch using BioLector and RoboLector (= microplate with automated sampling & feeding).



Glucose-xylose ratios, favoring specific strains, were tested in Fed-Batch conditions, achieving a stable co-culture with a 50% strain ratio.

3 TOWARDS METACOMMUNITIES

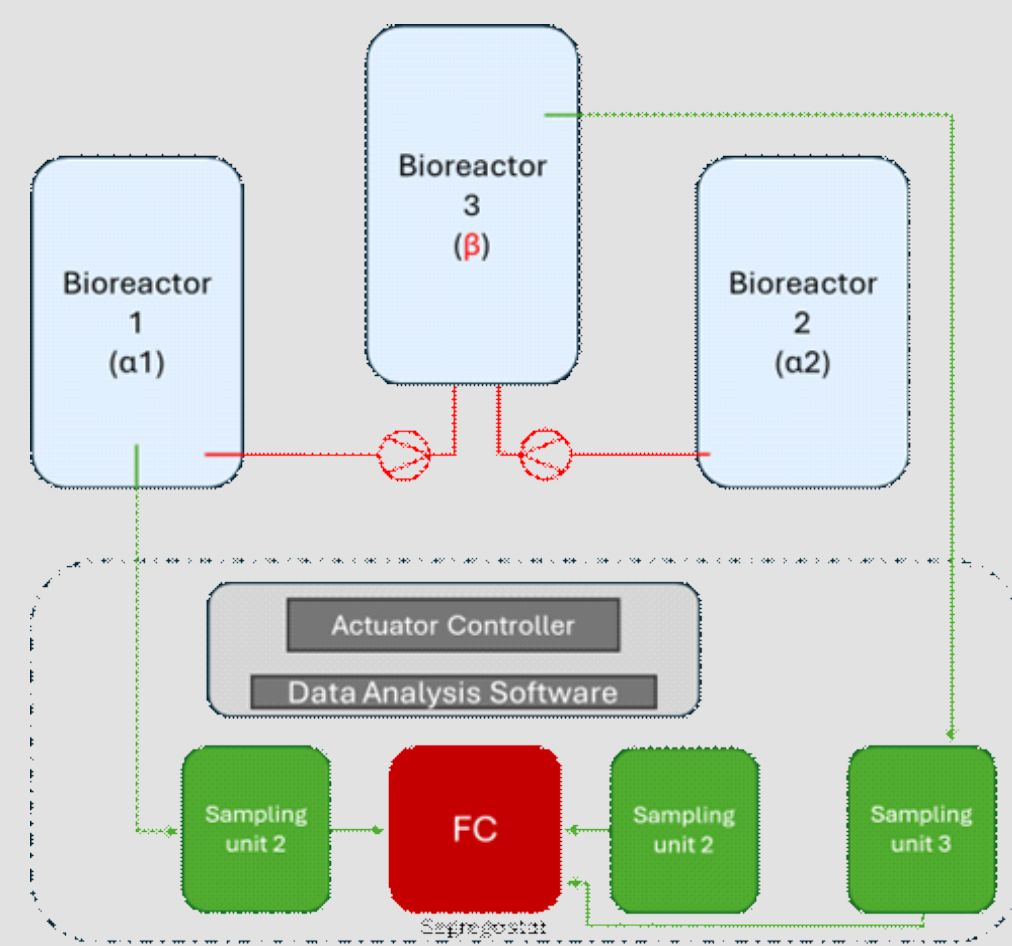
AAMN (Co-culture)

Continuous Culture of MetaCommunities

AAMN expands to interactions and abiotic parameters, with fingerprint development (cytometry/omics) to monitor microbial

Metacommunity

Spatial connection of CCMC enhances productivity, flexibility, and metabolic potential, integrating ecological concepts [4-5]



[1] Roell GW, Zha J, Carr RR, Koffas MA, Fong SS, Tang YJ. Engineering microbial consortia by division of labor. Microb Cell Fact [Internet]. BioMed Central; 2019;18:1–11. Available from: <https://doi.org/10.1186/s12934-019-1083-3>

[2] Martínez J, A, Bouchat, R., Gallet de Saint Aurin, T., Martínez, L. M., Caspeta, L., Telek, S., Zicler, A., Gosset, G., & Delvigne, F. (2025). Automated adjustment of metabolic niches enables the control of natural and engineered microbial co-cultures. Trends in Biotechnology. <https://doi.org/10.1016/j.tibtech.2024.12.005>

[3] Martínez JA, Delvenne M, Henrion L, Moreno F, Telek S, Dusny C, et al. Controlling microbial co-culture based on substrate pulsing can lead to stability through differential fitness advantages. bioRxiv [Internet]. 2022;2022.02.18.480836.

[4] Thompson, P. L., Guzman, L. M., de Meester, L., Horváth, Z., Ptácník, R., Vanschoenwinkel, B., Viana, D. S., & Chase, J. M. (2020). A process-based metacommunity framework linking local and regional scale community ecology. Ecology Letters, 23(9), 1314–1329. <https://doi.org/10.1111/ele.13568>

[5] Araujo, G., Montoya, J. M., Thomas, T., Webster, N. S., & Lurgi, M. (2024). A mechanistic framework for complex microbe-host symbioses. In Trends in Microbiology. Elsevier Ltd. <https://doi.org/10.1016/j.tim.2024.08.002>