AFT 2026

Advanced Fermentation Technology

BOOK OF ABSTRACTS

Polytech-Lille – Université de Lille

30 June – 1 July 2026

Strains Selection and Engineering

Advanced Fermentation Systems

Control strategies and Machine Learning

Autonomous Bioprocessing

Bioprocessing: From laboratory scale to Industry

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Welcome

Dear colleagues,

It is a great pleasure to welcome you to the first edition of **Advanced Fermentation Technology (AFT 2026)**, an international conference dedicated to the latest developments in fermentation science and technology.

AFT 2026 is held on 30 June and 1 July 2026 at Polytech Lille, University of Lille, France. This inaugural meeting brings together researchers, engineers and industrial professionals from around the world to exchange knowledge and discuss recent advances in fermentation technologies, digital twins, process modelling, translational bioprocessing and industrial biotechnology.

The ambition of AFT 2026 is to create a dynamic forum where scientific excellence meets technological innovation. Beyond the presentation of cutting-edge research, the conference aims to stimulate interactions between academia and industry, foster new collaborations, and encourage the exchange of ideas across disciplines and career stages. We are particularly pleased to welcome senior scientists, industrial experts, postdoctoral researchers, PhD candidates and early-career investigators to contribute to these discussions.

The scientific programme features oral communications selected from submitted abstracts, poster presentations, keynote lectures and dedicated networking opportunities. Together, these sessions provide a stimulating environment for sharing experiences, discussing emerging challenges and exploring future directions in fermentation technology.

We would like to express our sincere gratitude to all speakers, authors, sponsors, participants, members of the Scientific Committee, members of the Organising Committee, the VALOREEMA Industrial Chair, and the Adebiotech Association for their commitment and support. Their enthusiasm, expertise and contributions have been essential in making this first edition possible. We warmly welcome you to Lille and wish you an enjoyable and productive conference. We hope that AFT 2026 will inspire fruitful discussions, new partnerships and lasting scientific exchanges.

Prof. François Coutte
Chair of the Scientific Committee
Advanced Fermentation Technology 2026

Prof. Rozenn Ravallec
Chair of the Organizing Committee
Advanced Fermentation Technology 2026

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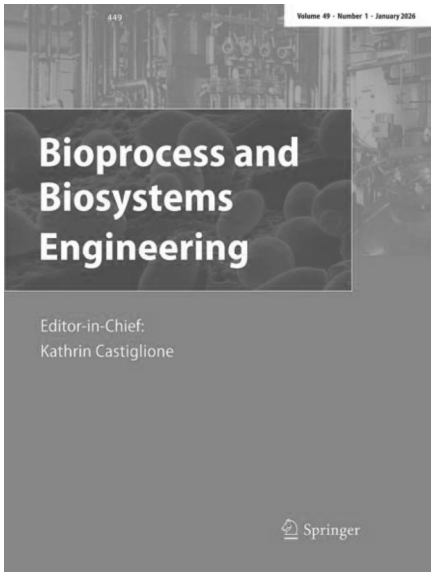

Editor-in-Chief: Kathrin Castiglione

Bioprocess and Biosystems Engineering

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- **Takes into account the environment and constraints of technical production processes**
- **Publishes original papers, short communications and invited critical reviews**

The journal Bioprocess and Biosystems Engineering is an international peer-reviewed forum for discussion between engineering and biological science to find efficient solutions in the development and improvement of bioprocesses.

The journal focuses attention on multidisciplinary approaches for integrative bioprocess design. Coverage includes rational manipulation of biosystems through metabolic engineering techniques to provide new biocatalysts. The journal examines model-based design of bioprocesses, such as up-stream processing, bioreactor operation and downstream processing, leading to new and sustainable production processes.

The journal offers new approaches for design of cellular systems, considering the environment and constraints of technical production processes, integration of recombinant technology and process design, as well as bioinformatics and process systems engineering.

The journal publishes original papers, short communications and invited critical reviews.

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Detailed programme

Day 1 – Scientific Advances and Technological Innovations

08:00 – 09:00 | Welcoming
09:00 – 09:30 | Opening Session

09:30 – 10:15 | Plenary conference : Christoph HERWIG (Körber Pharma, Austria)
The Potential of Digital Tools to Bridge Research Innovations to Viable Bioprocesses

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10:15 – 10:45 | Coffee break

10:45 – 12:30 | Session 1 – Selecting and Engineering Robust Strains for efficient fermentation
Chairs: François Coutte (University of Lille, France) & Philippe Jacques (University of Liège, Belgium)

Focus: *This session focuses on designing robust microbial strains adapted to real industrial constraints, including scale-up, downstream purification, and process integration. It highlights the use of techno-economic and model-guided frameworks combining metabolic, process, and cost modelling to guide early strain and process development. Key discussions will address how predictive and AI-enhanced models can support scalable, energy-efficient, and cost-competitive bioprocess design under market and regulatory constraints.*

10:45 - Talk 1.1 : Claus Lattemann (Lesaffre International, France)
From Strain Design to Manufacturing at Scale: Design principles and challenges bringing strains to commercial scale

11:10 - Talk 1.2 : Lisbeth Olsson (Chalmers University of Technology, Sweden)
Microbial robustness-a key to efficient bioprocesses

11:35 - Talk 1.3 : Ravinder Sharma (TetraPack, Sweden)
The make or break of precision fermentation: the choice of the host organism

11:50 - Talk 1.4 : Francesco Roppo (UMRt BioEcoAgro, Uni-Liège & Uni-Lille, Belgium)
Advancing Lignocellulosic Biomass Conversion into Lipopeptides through genetic engineering of B. subtilis guided by model-based constraint programming

12:05 - Talk 1.5 : Clément Dince (SEQENS, France)
Transitioning Toward Green Engineering: Precision Fermentation as a Sustainable Alternative to Chemical Peptide Synthesis — A Case Study of the PeptiCode™ Platform

12:20 - Flash poster presentation Session 1

12:30 – 14:00 | Lunch & Poster Session

14:00 – 16:00 | Session 2 – Advanced Fermentation Systems

Chairs: Nathalie Gorret (Toulouse Biotechnology Institute, France) & Petra Heidinger (Acib Gmbh, Austria)

Focus: *This session will highlight advances in fermentation system design that enable a robust transition from laboratory to industrial scale. It will address innovations in bioreactor technologies, automation, and process intensification, with a strong focus on scale-up methodologies and early techno-economic evaluation. Discussions will center on how integrated engineering and economic tools can improve productivity, reliability, and cost efficiency across diverse fermentation platforms.*

14:00 - Talk 2.1 : Petra Heidinger (Acib GmbH, Austria)

From CO_2 to Protein: Advancing Pilot Scale Gas Fermentation with C. necator

14:25 - Talk 2.2 : Nico Cruz Bournazou (TU Berlin, Germany)

Experimental Design and Operation Strategies for Accelerated Bioprocess Development

14:50 - Talk 2.3 : Pedro Ferrandiz (Genodics, France)

New tools to optimize bioprocesses

15:05 - Talk 2.4 : Elodie Choque (Université Picardie Jules Verne, France)

Optimization of semi-solid fermentation of agricultural by-products using Aspergillus tubingensis for a without waste use

15:20 - Talk 2.5 : Stephane Guillouet (Toulouse Biotechnology Institute, INSA, France)

Bioreactor Engineering for Bacterial Production of Biomolecules from CO_2

15:35 - Talk 2.6 : Nabila Imatoukene (URD ABI, AgroParisTech, France)

Enhanced de novo Ferulic Acid Production in Biphasic Yeast Fermentation

15:50 - Flash poster presentation Session 2

16:00 – 16:30 | Coffee break & Poster Session

16:30 – 18:30 | Session 3 –Control Strategies, Machine Learning and Digital Twins for Bioprocess Design

Chairs: Frank Delvigne (University of Liège, Belgium) & Nico Cruz Bournazou (TU Berlin, Germany)

Focus: *This session will focus on sensors, process analytics, and data-driven control strategies enabling real-time monitoring and optimization of fermentation processes. It will highlight the role of AI, hybrid models, and digital twins in improving process robustness, scale-up predictability, and decision-making across the bioprocess lifecycle. Key discussions will address model reliability, techno-economic trade-offs, and the validation of digital control frameworks under industrial constraints.*

16:30 - Talk 3.1: Moritz Von Storsch (DataHow, Switzerland)

How digital tools can support scale-up: Using hybrid modeling and transfer learning for across scale predictions

16:55 - Talk 3.2 : Alex Fedorec (University College of London, United Kingdom)

Reinforcement learning for the control of microbial communities in bioreactors

17:20 - Talk 3.3 : Caroline Solon (ATV Technologies, France)

Smart fermentation at scale: How CDMOs accelerate bioprocess development through AI and Digital Twins

17:35 - Talk 3.4 : Jean-Sébastien Guez (Institut Pascal, Université de Clermont, France)

Deep Learning Unlocks Real-Time Morphology Based Characterization of Cell Populations in Bioreactors

17:50 - Talk 3.5 : Gregory Batt (INRIA, Institut Pasteur, France)

Using combinatorial cloning approaches and automated cytometry to optimize bioproduction in yeast

18:05 - Talk 3.6 : César Rodriguez-Fano (Université de Claude Bernard Lyon 1, France)

Real-time monitoring of Escherichia coli cultures using multiple spectroscopic methods

18:20 - Flash poster presentation Sessions 3

18:30 – 21:00 | Poster Session (Cocktail)

Day 2 – Translational Research and Industrial Collaboration

08:15 – 09:25 | Session 4 – Autonomous Bioprocessing for Industrial Scalability

Chairs: Marcin Lukaszewicz (University of Wrocław, InventionBio, Poland) & Julien Boutet (Seqens, France)

Focus: *This session will focus on the transition toward autonomous and continuous bioprocessing through the integration of upstream fermentation and downstream purification. It will highlight process design, control, and integration strategies that enhance productivity, robustness, and resource efficiency. Discussions will emphasize the role of techno-economic evaluation in guiding decisions from pilot to full-scale industrial operation.*

08:15 - Talk 4.1 : Anna-Lena Heins (University of Hambourg, Germany)

How much do we need to monitor and control for development of scalable, high-yielding and robust bioprocesses?

08:40 - Talk 4.2 : Enver Felix Loayza Mora (INBM TUB Freiberg, Germany)

Droplet-based microfluidics and automation to overcome challenges in the screening of secondary metabolites during bioprocessing

08:55 - Talk 4.3 : Eric Hiller (University of Hohenheim, Germany)

Model-based process design for surfactin production with Bacillus subtilis

09:10 - Talk 4.4 : Yasmine Jerad (iMEAN, France)

Digital Twins in Modern Fermentation - Integration of high-predictive models in Autonomous Fermentation

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9:25 -10:10 | Industrial sponsor presentation (5 minutes/sponsor)

Chair : Rozenn Ravallec (University of Lille, France)

10:10 – 10:40 | Coffee break

10:40 – 12:25 | Session 5 – Translational Bioprocessing: From laboratory scale to Industry

Chairs: Anthony Bressin (AnBreiL, France) & Elodie Wattez (Ingredia, France)

Focus: *This session will address how to translate laboratory-scale innovations into industrial bioprocesses by aligning scalability, regulatory compliance, and economic viability. It will highlight technology transfer models, pilot and demo-scale infrastructures, and sector-specific constraints across food, pharma, green chemistry, and bio-based materials. Key discussions will focus on overcoming technical and regulatory bottlenecks through stronger collaboration between academia, industry, and regulators.*

10:40 - Talk 5.1: Bardor Murielle (Algae Biologics, University of Rouen, France)

From Fundamental research regarding the protein N-glycosylation pathways in microalgae to the production of microalgae-made biologics at industrial scale

10:55 - Talk 5.2 : Ildar Nisamedtinov (Lallemand, Estonia)

From ideation to execution: essential steps in industrial bioprocess development

11:10 - Talk 5.3 : Vincent Usache (Microphyt, France)

Microphyt - From Lab to Market: Scaling Microalgae Production and Extracts for Industry

11:25 - Talk 5.4 : Ghislain Sanhaji (ARD, France)

From Innovation to Production: Mastering the Art of Industrial Scale-up to Secure Your Path to Market

11:40 - Talk 5.5 : Tambi Kar (Galactic, Belgium)

Industrial deployment of recycled-carbon fermentation: A translational framework for low-emission bioprocesses

11:55 - Talk 5.6 : Olivier Galy (TWB-INRAE, France)

Adoption of miniaturized an automated approaches to assist R&D for bioprocess developments

12:10 Talk 5.7 : Lilian Favacho Rodrigues (B4C, France)

Bioindustry 4.0 : Leveraging advanced digital solutions to empower the European bioindustry sector

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12:25 – 13:00 | Round table Discussion

Theme: *This roundtable will explore how the convergence of systems biology, data science, artificial intelligence, equipment and bioprocess engineering can redefine the future of fermentation and bioproduction. In addition to scientific and technological challenges, the discussion will address the evolving economic landscape and funding environment for breakthrough biotechnology projects, particularly in fermentation-based innovations*
Chairs: Jean-Pierre LEAC (SATT Nord), Lionel Genetelli (Univ- Lille)

Contributors : Christoph Herwig (Körber Pharma, Austria); Michael Krel (Sofinnovpartners, France); Pierre Monsan (Toulouse Biotechnologie Institute, France); Guillaume Boissonnat (Pili, France); Christophe Luguel (B4C, France) ; Etienne Vervaeck (NSL, France)

13:00 – 13:30 | Closing session and Poster Prices

13:30 – 14:30 | Networking Lunch

14:30 – 16:30 | Pôle Universitaire d'Innovation Lille (PUI) event & Lab visit

Conclude the congress with a convivial moment to learn more about opportunities for collaboration with public research alongside the business development team of the PUI Lille.

14:30-14:45 |Presentation of PUI Lille

14:45-15:15 |Coffee break & networking

15:15-16:30 | Lab tours of BioEcoAgro : Biopesticide production : from cell to bioprocess

Oral presentation

Plenary conference : The Potential of Digital Tools to Bridge Research Innovations to Viable Bioprocesses

HERWIG Christoph[a]

[a]Lisalis GmbH, Jenbachgasse 73-2, 1130 Vienna, Austria
[b]Körber Pharma Austria GmbH, Mariahilferstrasse 88A, 1070 Vienna, Austria

Abstract:

Data is the new gold. Also in the biotech domain, we have to enrol into using the data, else our business will not survive. But how? What are the steps to take in order to cope with the vast variety of data sources? Which steps do we need to do to preprocess and contextualize the data, so that we can convert data into information and knowledge?

However, we shall not do all this without having clear use cases in mind. This contribution also focuses on the potentials of using well contextualized data to develop digital twins and demonstrated the potential to deploy the digital twins along the product life cycle.

We show how Physics Informed AI enables the deployment of digital twins in real time use cases, as well as how digital twins can be concatenated to build end-to-end digital twins. The latter one allows in addition robust prediction of product quality at the end of the process chain as well as compensatory and optimizing actions to avoid batch failure and increase business economy.

Potentials of the show approach are: Quicker process development and characterization, more predictive in tech transfer and validation, and allow for process robustness and optimization in manufacturing using digital twin based control as well as real time release using end to end solutions.

Keywords: Digital Twins, Digital Transformation, Process Knowledge

T1.1 : From Strain Design to Manufacturing at Scale: Design principles and challenges bringing strains to commercial scale

LATTEMANN Claus[a]

[a]Lesaffre Institute of Science and Technology, Lesaffre International, 101 Rue de Menin, 59700 Marcq-en-Baroeul - France

Abstract :

Robust fermentation processes operating at industrial scales beyond 100 m³ are prerequisites for a successful bioeconomy meeting economic and sustainability targets. Translating bioprocesses from laboratory-scale operations—which operate under controlled, homogeneous conditions—to industrial-scale bioreactors presents significant challenges. Industrial bioreactors are characterized by substantial spatial gradients in substrate concentration, dissolved oxygen, pH, and temperature. These heterogeneous conditions impose significant metabolic burden on production strains, leading to reduced productivity and/or quality compared to bench-scale performance.

Effective strain design for manufacturing scale must address these challenges through concurrent development with process engineering. Strain optimization coupled bioprocess design through iterative cycles aligning strain characteristics with bioreactor design and operating strategies and process constraints guiding strain engineering targets allows mitigating scale-up risks.

Future success can be enabled through predictive computational models linking strain genotype to bioprocess performance, enabling rational co-optimization of strain and manufacturing conditions for robust, scalable production systems meeting the demands of a sustainable bioeconomy.

Keywords : Scale-up / scale-down, bioprocess engineering, large scale manufacturing

T1.2 : Microbial robustness-a key to efficient bioprocesses

OLSSON Lisbeth *,[a], GUERRA GIACON Thamiris, TORELLO PIANALE Luca[a] , TRIVELLIN Cecilia[a], VILELA Nathália[a]

[a]Industrial Biotechnology, Department of Life Sciences, Chalmers University of Technology

Abstract:

Yeast is broadly exploited for industrial use, and strains are constantly improved to meet the requirements to produce the targeted product with high yield, productivity and titer. When scaling up bioprocesses towards industrial scale, it is not uncommon that strains perform poorer than in the laboratory. Successful strains have consistent performance also in presence of different perturbations such as those encountered during scale-up, i.e. their performance is robust. The molecular trait behind microbial robustness is important to understand to further elaborate of the potential of different cell factories.

To allow investigation on microbial robustness in face of different perturbations we have developed a toolbox as a fundament to bring deeper insights. A method to quantitatively assess microbial robustness has been developed. This method allows a high throughput evaluation, in a perturbation space where different cellular function can form the basis for the evaluation. Another important approach has been the examination of intracellular status in face of perturbations using biosensors, for instance intracellular ATP and pH.

In this presentation some applications of microbial robustness will be demonstrated,

(i) studies adaptive laboratory evolution processes to gain information of determinative factors of robustness.

(ii) using published data set to identify genetic markers for robustness.

(iii) studies of the influence of contaminants in bioethanol production and their influence on yeast robustness

Keywords: quantification, biosensor, adaptive evolution, genetic markers, bioprocess contaminants

T1.3 : The make or break of precision fermentation: the choice of the host organism

SHARMA, Ravinder[a],CAMPOS, Joana[a*], GRENOV, Daniel[a]

[a]Tetra Pak Processing Equipment AB, Lund, Sweden

Abstract :

The selection of an appropriate host organism for recombinant protein production in the food industry requires balancing process robustness with efficient product recovery. While intracellular expression in fast-growing hosts such as bacteria offers short development and cultivation timelines, downstream processing often necessitates cell disruption, increasing cost and complexity. In contrast, organisms with a natural capacity for protein secretion, including filamentous fungi, can simplify product recovery but introduce significant challenges. Their slower growth rates markedly extend strain development and cultivation times, and their complex physiology demands tight process control to prevent sporulation and secondary metabolite formation, including mycotoxins. Compared to filamentous fungi, yeast-based systems offer a compromise by enabling secretion within a eukaryotic context while retaining growth kinetics and process control closer to bacterial platforms. Nevertheless, maintaining strictly controlled conditions to ensure product consistency often compromises overall process robustness and scalability. This balance remains a central constraint in developing economically viable recombinant protein production platforms for food applications.

Keywords : Host, Precision Fermentation, Robustness

T1.4 :Advancing Lignocellulosic Biomass Conversion into Lipopeptides through genetic engineering of *B. subtilis* guided by model-based constraint programming

Francesco Roppo[1,2], Nastassia Kaugarenia[1], Joachim Niehren[3], Florian Chemarin[1], Sigrid Gorgen[2], Matthieu Duban[1], François Coutte[1], Philippe Jacques[2]

[1] *Université de Lille, UMRt 1158 BioEcoAgro, Polytech-lille, Villeneuve d'Ascq, France.*
[2] *Université de Liège, UMRt 1158 BioEcoAgro, Centre Terra, Gembloux, Belgium*
[3] *Cristal laboratory, Inria, Université de Lille, Villeneuve d'ascq, France*

Lipopeptides produced by *Bacillus subtilis* have emerged as sustainable alternatives to their synthetic counterparts, offering superior biodegradability and reduced environmental impact. However, the high cost of substrates continues to pose a major challenge to their commercial viability. To address this issue, genome optimization of the production strain is essential for developing efficient microbial chassis for new bioprocesses. This study aims to boost the valorization of cost-effective renewable agro-industrial by-product by developing new *B. subtilis* cell-factories through the application of a biocomputational tool.

The hydrolysis of lignocellulosic biomass yields a xylose-rich substrate that cannot be efficiently converted into lipopeptides due to intracellular bottlenecks. By modeling *B. subtilis* metabolism, and using constraint-based programming, multiple non-redundant candidate genes were identified for genetic modification at different levels (carbon, amino-acids metabolism, etc). The *in-silico* computation was verified by wet-lab experimentation. This approach leads to enhanced carbon uptake and optimized intracellular carbon flux resulting overall in increased lipopeptide production in xylose-rich media. These findings provide a foundation for time-efficient and knowledge-driven optimization of *Bacillus* strains for the valorization of renewable resources.

Keywords: Metabolic Engineering, Biotechnology, Bacillus, Lipopeptides

References:

[1] L. A. Sarubbo et al., 'Biosurfactants: Production, properties, applications, trends, and general perspectives', *Biochemical Engineering Journal*, vol. 181, p. 108377, Apr. 2022 [Online]. Available: 10.1016/j.bej.2022.108377.

[2] M. Klimek-Szczykutowicz et al., 'Bioferments and Biosurfactants as New Products with Potential Use in the Cosmetic Industry', *Applied Sciences*, vol. 14, no. 9, p. 3902, May 2024 [Online]. Available: 10.3390/app14093902.

T1.5: Transitioning Toward Green Engineering: Precision Fermentation as a Sustainable Alternative to Chemical Peptide Synthesis —A Case Study of the PeptiCode™Platform

DINCE Clément[a], PALLAS Stéphanie[a], and TISSOT-LECUELLE Ghislaine[a]*

[a]SEQENS Biotechnologies, 21 chemin de la Sauvegarde, 69130 Ecully, France

Abstract:

The personal care industry is currently undergoing a paradigm shift, driven by consumer demand for high-performance ingredients that align with rigorous naturality and sustainability standards[1]. Consequently, cosmetic ingredient suppliers are strategically pivoting toward production methodologies that utilize renewable carbon sources and "cleaner" processes, such as biocatalysis, precision fermentation and green engineering[2].

Peptides, while central to modern cosmetic efficacy, present a significant sustainability challenge. Historically, these molecules were synthesized via chemical synthesis, such as Solid Phase Peptide Synthesis (SPPS)—a process characterized by heavy solvent consumption, the use of PFAS, and substantial waste generation, often accompanied by diminishing yields as sequence length increases[3].

To address these limitations, SEQENS Biotechnologies developed PeptiCode™, a biotechnological platform leveraging precision fermentation and green engineering. This sustainable platform enabled the production of the first European-made, biotech Hexapeptide-9—a key bioactive sequence found in collagen types IV and XVII. This presentation will detail the microorganism genetic engineering, the upstream process optimization, and the specific downstream strategies required to achieve high peptide purity and yields.

Keywords : Peptide, Precision Fermentation, Downstream-Process, Sustainability, Green Engineering

References :

1) Alviri H, Lynes J, Habib K. (2025). Beyond green chemistry: a comprehensive review of how sustainability has been integrated into cosmetic research. *Global Sustainability 8*, e16, 1-18. doi:10.1017/sus.2025.5
2) Harasim, E., et al. (2026). Sustainable utilization of natural plant resources in eco-friendly cosmetic production. *Journal of Ecological Engineering, 27*(2), 53-68. doi.org/10.12911/22998993/210719
3) Akbarian M, Khani A, Eghbalpour S, and Uversky VN (2022) Bioactive Peptides: Synthesis, Sources, Applications, and Proposed Mechanisms of Action. *Int J Mol Sci.* 23(3):1445. doi: 10.3390/ijms23031445.

T 2.1 : From CO_2 to Protein:

Advancing Pilot Scale Gas Fermentation with *C. necator*

HEIDINGER Petra

acib GmbH – Austrian Centre of Industrial Biotechnology
Krenngasse 37, 8010 Graz, Austria

Abstract :

The microbial production of single-cell protein (SCP) through gas fermentation with *Cupriavidus necator* offers a sustainable way to decouple food and feed supply from conventional agricultural constraints. Using CO_2 as an unconventional, currently underutilized feedstock presents a promising opportunity to further separate protein production from both fossil and land-based resources. However, scaling these processes to the pilot level is limited by suboptimal gas–liquid mass transfer of the H_2 and O_2 substrates. To address these challenges, a novel reactor design that enhances gas transfer through elevated pressure and tailored gas compositions was developed. Our results show that this design significantly increases volumetric productivity compared to conventional gas fermentation systems. Pilot-scale experiments demonstrate substantially higher biomass yields, enabled by improved gas transfer performance and an optimized feeding strategy. Together, these results bridge biological potential and engineering implementation, outlining a realistic path towards industrial-scale CO_2-derived protein production within a circular bioeconomy.

T 2.2 : Experimental Design and Operation Strategies for Accelerated Bioprocess Development

Cruz Bournazou, M. Nicolas[a], Ernesto Martinez[a], Peter Neubauer[a]

[a]KIWI-biolab, Chair of Bioprocess Engineering, Technische Universität Berlin, Ackerstrasse 76, 13355, Berlin, Germany

Abstract:

Industrial-scale bioprocesses are inherently dynamic and spatially heterogeneous, exhibiting temporal variability and population heterogeneity that pose significant challenges during scale-up. To address this, traditional scale-down approaches aim to capture large-scale effects qualitatively but often fail to reproduce the full dynamic cellular perspective encountered in industrial reactors.

We present recent advances in the integration of computational tools for experimental design and operation, laboratory automation, and data management aimed at accelerating bioprocess development. Machine learning augments traditional modeling by automating model construction, experimental planning, and decision-making across the experimental cycle. Bayesian experimental design prioritizes informative experiments under uncertainty, and reinforcement learning enables online redesign of dynamic experiments to maximize information gain despite biological variability, hardware faults, and noisy measurements. We also discuss a telescopic model-based design-of-experiments methodology that focuses on reducing uncertainty in key performance indicators (KPIs) at predicted optima rather than minimizing parameter uncertainty, thereby accelerating identification of economically favorable operating conditions at industrial scale. Combined with scale-down experimental techniques, these approaches enable significantly more reliable scale-up.

Finally, the orchestration of autonomous workflows requires scalable, knowledge-oriented data infrastructures that automatically capture semantically rich metadata, ensure provenance and traceability, and enable information transfer across scales and projects. Collectively, these mechanistic, data-driven, and ML-enabled advances provide a roadmap for reproducible, faster, and more robust bioprocess development and industrial implementation.

Keywords: High Throughput Bioprocess Development, Machine learning and Lab automation, Optimal Experimental Design, Reinforcement learning, Knowledge-driven data infrastructure

T 2.3 : A New Tool for Optimizing Microorganism cultures, with Biological Music

PREVOST LOUKIANOV Victor[a] and FERRANDIZ Pedro[a]

[a]GENODICS SAS, 23 rue Jean-Jacques Rousseau 75001 Paris

Abstract :

The Genodic process is the result of more than 30 years of R&D: it is an innovative method of regulating protein biosynthesis using acoustic frequencies. The scientific basis relies on the European patent of French physicist Joël Sternheimer (EP0648275B1) relating to the "epigenetic regulation of protein biosynthesis by scale resonance."

The process has been implemented in agriculture since 2008 by the company Genodics: 300 installations at 150 farms across 2,500 hectares have made it possible to optimize yields and quality, reduce the impact of biotic and abiotic stress, and reduce inputs. The process has been tested by the ERRMECe laboratory at CYU on the production of the dehydrin protein in pea crops under water stress conditions (CYU, 2021).

Proof of concept was demonstrated on the bioluminescent bacterium Vibrio f. (University of Metz, 1992–95), on Saccharomyces c. yeast in bread making (ENSMIC, 1993), in winemaking on alcoholic fermentation and the production of veil on yellow wine (Genodics 2009–2012), on malolactic bacteria in winemaking (Genodics 2010–2013), on cell cultures (with JURKAT and SKOV3 lines, respectively) on the production of Interleukin 2 (ESPCI, 2003) and Integrin α5-GFP (CYU, 2021).

Here we present these studies in detail, along with the prospects they open up for all types of fermentation and microbial production processes or those using eukaryotic cell cultures.

Keywords : proteodies, biosynthesis, bioacoustics, epigenetics, microorganisms

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T 2.4 Optimization of semi-solid fermentation of agricultural by-products using *Aspergillus tubingensis* for a without waste use

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Abstract :

The bioconversion of agricultural byproducts is a hot topic: (i) their bioconversion via bacterial fermentation for the production of lactic acid, PHA, or for the production of active antioxidant or antimicrobial metabolites, (ii) their bioconversion via colonization by edible fungi for their production or the production of mycomaterials for design and/or construction, (iii) their bioconversion for the production of insects, a protein-rich alternative food source to meat. However, research on the valorization of these agro-resources through semi-solid fermentation using filamentous fungi is more limited in the literature. During this presentation, we will present the results obtained for the valorization of two agri-food byproducts: chicory root (Rambaud et al., 2025) and brewer's spent grain. We will first discuss the optimization of the semi-solid fermentation process of a GRAS filamentous fungus, *Aspergillus tubingensis*, based on the composition of each of these resources (Choque et al., 2015; Lopez de Leon et al., 2019). We will then demonstrate that, following fermentation, a pressing step enables the recovery of both the antioxidant-rich fermentation juice and the solid residue—composed of fermented agri-food residues and fungal biomass—for various applications: the extraction of fungal chitin; the production of bioactive films for cosmetic use; and the production of construction materials (Choque et al., 2021; Rezzani et al., 2022).

Keywords: agro-food waste valorization ; semi-solid fermentation ; antioxidant ; mycomaterials ; chitosan

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T 2.5 : Bioreactor Engineering for Bacterial Production of Biomolecules from CO_2

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Abstract :

The development of Carbon Capture, Storage, and Utilization (CCUS) processes using microorganisms is one of the promising technologies for reducing greenhouse gas emissions. The use of microorganisms naturally capable of transforming CO_2 for their growth and the production of biomolecules will enable the development of alternative processes for producing fuels and chemicals from industrial gas effluents.

Through a multidisciplinary strategy combining metabolic engineering and biochemical engineering, we have developed *Cupriavidus necator* as a platform for producing biomolecules from CO_2. In parallel, we designed a range of culture systems at different scales (flasks, 500 mL to 10-liter gas bioreactors) to ensure autotrophic growth under safe conditions among them a high-pressure CSTR gas bioreactor and a gas bioreactor coupled to a Hollow Fiber Membrane Contactor.

During this presentation, we will illustrate the development of bioreactors with examples of growth and production performance evaluations of our engineered *C. necator* strains in our various bioreactors. This work has enabled us to significantly improve the production performance of molecules such as isopropanol in quantities exceeding g/L from CO_2.

Keywords : Carbon Capture Storage and Utilization, Gas fermentation, Cupriavidus, CO_2, Hollow Fiber Membrane Contactor

T2.6 : Enhanced de novo Ferulic Acid Production in Biphasic Yeast Fermentation

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Abstract :

Ferulic acid (FA) is a phenolic compound with diverse biological properties, widely used in the food and cosmetic industries[1–3]. Its production from fermentation is a promising strategy because its extraction from biomass is costly[4]. The objective of this study was to produce FA from D-glucose using an engineered *Saccharomyces cerevisiae* strain, with the goal of developing a more economically viable medium and fermentation process. Given the hydrophobic nature of FA, which limits its accumulation in the aqueous phase, an appropriate bioprocessing approach was implemented, involving the cultivation of *S. cerevisiae* using both batch and fed-batch biphasic fermentation strategies.

Our results showed that fed-batch biphasic fermentation system resulted in almost a two-fold increase in FA production compared to batch one (312.6 $mg.L^{-1}$ and 176.7 $mg.L^{-1}$, respectively). Regarding the distribution of FA and its related intermediates, such as *p*-coumaric acid (*p*-CA) and caffeic acid, during batch and fed-batch fermentations, the acidic pH facilitated the migration of *p*-hydroxycinnamic acids (*p*-HCAs) predominantly into the organic phase, enabling effective detoxification of the broth and efficient recovery of *p*-HCAs. Increased solubility of *p*-HCAs in the fatty alcohol phase can be attributed to their protonation when the pH is lower than their respective pK1 values, which are 4.65, 4.62, and 4.56, for *p*-CA, caffeic acid, and FA, respectively.

Keywords Ferulic acid, medium optimization, *Saccharomyces cerevisiae*, biphasic fermentation

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T 3.1: How digital tools can support scale-up: Using hybrid modelling and transfer learning for across scale predictions

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Abstract :

Scale-up remains a persistent challenge in upstream process development. While methodologies have evolved from maintaining constant dimensionless numbers to more advanced approaches, such as characterizing gradients via computational fluid dynamics and developing physical scale-down models, reliably predicting large-scale process performance from small-scale data for new processes is still difficult.

In this work, we propose a framework that combines hybrid modeling with transfer learning to predict large-scale performance. The approach leverages (i) small-scale experimental data from the process under development and (ii) historical datasets from other processes operated across multiple scales.

Our results indicate that large-scale performance can be predicted with high accuracy when the new process exhibits similarity to processes included in the training data. This finding is particularly relevant for platform process formats widely used in the pharmaceutical industry, where either reliable large-scale prediction can be achieved directly, or knowledge gaps can be systematically identified and addressed through targeted design-of-experiments strategies.

Keywords : Hybrid modelling, Transfer Learning, Scale-up, Optimization

T 3.2 : Reinforcement learning for the control of microbial communities in bioreactors

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Abstract :

Engineered microbes have the potential to revolutionise sectors including food, manufacturing, health, and climate. However, our current reliance on engineering monocultures limits their application and impact. Through division-of-labour, synthetic communities can provide the solution to monoculture limitations but producing predictable, functional, and compositionally robust communities remains a challenge that has stifled their more extensive use. Here we demonstrate the efficacy of an artificial intelligence for the control of co-cultures within continuous bioreactors. We show, *in silico*, that a reinforcement learning agent is able to learn to accurately control simple microbial communities through perturbing the bioreactor in which they are growing. Using bang-bang control of properties such as dilution and feed-composition, we can stabilise communities at target densities or ratios, guide community composition through pre-designed trajectories, or optimise the yield of measurable outputs. In particular, the reinforcement learning agent outperforms classical control approaches when faced with infrequent sampling. Although reinforcement learning requires a significant amount of data to train, we demonstrate that good control policies can be learned online, within 24 hours, by using parallel bioreactors. Together, these results establish reinforcement learning as a powerful and practical framework for enabling reliable, adaptable, and scalable control of synthetic microbial communities.

Keywords : Reinforcement learning control; Continuous bioreactors; Division-of-labour

Références :

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T 3.3 : Smart fermentation at scale: How CDMOs accelerate bioprocess development through AI and Digital Twins

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Abstract:

AI: Artificial Intelligence or Accelerator of Innovation and Accelerated Industrialization? A 3-in-1 paradigm evolving beyond mere mathematical modelling to become a real-time copilot for smarter fermentation. Fermentation processes are already monitored and controlled; AI improves the precision and timeliness of actionable indicators, enabling proactive interventions. At ATV, this is illustrated through 3 collaborative use cases supporting faster bioprocess development and more robust scale-up. A first example relies on IoT-connected UV probes (BioOptics+), which provide continuous, non-intrusive monitoring of biological states. Combined with machine-learning models from BioIntelligence Technologies, these online signals enable earlier detection of deviations and support data-driven decision-making. Integrating AI into continuous fermentation has been shown to maintain stable operations, accelerate time-to-market, and improve productivity under real production constraints. Finally, ATV has developed an internal digital twin that learns process dynamics from online data to anticipate behaviour, paving the way toward autonomous control, lower operating costs, and a smaller environmental footprint. Together, these examples show how AI and digital twin technologies can accelerate the industrialization and market deployment of innovative technologies for next-generation biomanufacturing.

Keywords: Real-Time Decision Support, Early Deviation Detection, Online Monitoring, Continuous Bioprocessing, Predictive Digital Twin

T 3.4 : Deep Learning Unlocks Real-Time Morphological Characterization of Cell Populations in Bioreactors Equipped with *In Situ* Microscopy

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Abstract:

Biotechnological production processes involving living cells are frequently affected by undesirable physiological states that reduce both productivity and product quality. In bioreactors, cells experience heterogeneous microenvironments caused by hydrodynamic stress and spatial gradients in dissolved gases, pH, nutrients, and metabolites, effects that become more pronounced at industrial scale. Conventional process parameters provide only population averaged information and fail to capture extrinsic sources of cellular heterogeneity, despite their well-established impact on process performance (Delvigne et al., 2014). This underscores the need for advanced monitoring strategies capable of linking culture conditions to single cell behaviour in real time. To address this limitation, a Convolutional Neural Network-based framework (YOLO) was developed to enhance the image analysis process of *in situ* microscopy images and deliver real time morphometry measurements of cell populations during bioprocessing. The first application focused on the monitoring of mammalian cell in bioreactors for biopharmaceutical production, enabling real-time estimation of cell density and viability. The system also discriminated multiple cellular phenotypes and detected a distinct “bulged” morphology associated with fluctuating substrate availability, providing a potential early indicator of suboptimal culture conditions (Guez et al., 2023). In a second application, morphological changes of anaerobic thermophilic archaea during a biomethanation process in a bubble column were explored. This process allows to produce methane (CH_4) from hydrogen (H_2) and carbon dioxide (CO_2), which is considered as a technology “next” to move away from the current system of energy production based on fossil fuels towards intermittent renewable energies (Biderre-Petit et al., 2024).

Keywords: PAT tool, Deep Learning, Image Analysis, Bioprocess Engineering, Heterogeneity

References

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T 3.5 : Using combinatorial cloning approaches and automated cytometry to optimize bioproduction in yeast

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Abstract:

Small-scale, low-cost bioreactors are emerging as powerful tools for microbial systems and synthetic biology research. Sophisticated experiments can be performed with high reproducibility. However, existing setups have limited in situ measurement capabilities. Researchers usually need to manually extract, process and measure culture samples using sensitive and specialized instruments. Manual interventions strongly **constrain** the available temporal resolution and reactiveness capabilities.

In this talk, I will present ReacSight, a generic strategy to enhance bioreactor arrays with automated measurements capabilities and reactive experiment control. It can be used to enhance any computer-controlled plate-based measurement device with pipetting capabilities and automation. ReacSight leverages the affordable Opentrons pipetting robots. It is ideally suited to integrate open-source, open-hardware components but can also accommodate GUI-only components. Applications include the control of an artificial differentiation system in yeast to create consortia with tuneable composition, and the characterization of protein secretion under various stress conditions to optimize production in yeast. I will conclude with a short discussion on ongoing research directions.

Joint work with past and present InBio team members.

Keywords: lab automation, optimization of protein secretion

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T 3.6 : Real-time monitoring of *Escherichia coli* cultures using multiple spectroscopic methods

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Abstract:

Real-time monitoring of fermentations is essential to address the challenges of cost, productivity, and quality in the biotechnology industry[1]. While real-time methods are well-established for pH and dissolved oxygen, monitoring of biological parameters and chemical compounds still largely relies on discrete and time-consuming measurements[2]. Spectroscopic methods have emerged as promising, rapid and non-destructive alternatives to obtain continuous information on these variables[3]. In this study, various spectroscopic methods - dielectric, Raman, excitation-emission matrix (EEM) fluorescence and ^{1}H nuclear magnetic resonance (NMR) - were evaluated for their potential to monitor *Escherichia coli* aerobic growths in a 5L-bioreactor. Dielectric spectroscopy, combined with linear or PLS modelling, allowed to monitor biomass in calibration batches but showed reduced predictive performances on independent batches. Raman and NMR spectroscopy enabled glucose and biomass monitoring, and also by-products for NMR. Interestingly, NMR spectra required mostly simple linear modeling (except for biomass), whereas Raman spectra analysis always relied on PLS modeling. EEM fluorescence spectroscopy revealed fluorescence increase during cultures at different combinations of emission-excitation wavelength likely related to biofluorophores. Unfolded EEM spectra combined with PLS modelling enabled the monitoring of biomass and glucose. Overall, the implementation of these sensors could reduce the time and cost associated with offline measurements. Their effective combination could also enable better understanding and adaptive control of bioprocesses. More work is planned to further study the potential and the robustness of each technique, and also the reliability of the built models.

Keywords: Bioprocess, real-time monitoring, spectroscopic methods, MVDA (MultiVariate Data Analysis), PLS (Partial Least Squares)

References:

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T 4.1 : How much do we need to monitor and control for development of scalable, high-yielding and robust bioprocesses?

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Abstract :

To address the challenges posed by climate change and the demand for sustainable production, robust and efficient bioprocesses are alternatives to conventional chemical processes. However, many promising bioprocesses that perform well at laboratory scale, fail upon scale-up to industrial scale. This discrepancy amongst other arises from environmental gradients and mixing insufficiencies in large bioreactors causing phenotypic population heterogeneities, initiating stress responses, enhancing metabolic burden and genetic instability. Then traditional assumptions of homogenous behaviour of isogenic cells under well-mixed conditions are challenged by complex, dynamic intracellular networks and spatial-temporal environmental variations, necessitating a multi-level bioprocess monitoring approach instead of traditional bulk measurement.

Here, the relevance of bioprocess monitoring strategies across hierarchical cell-physiological levels critical for bioprocess understanding and control are discussed. Measurement techniques are highlighted from inline sensors enabling real-time control to offline comprehensive analyses, and contrasts laboratory-scale flexible, multi-level monitoring with industrial-scale demands for robust, cost-effective, and regulative-compliant systems. Particular emphasis is placed on the integration of single-cell analysis methods, which enable direct assessment of phenotypic population heterogeneity gaining essential insights into amongst other metabolic states and stress responses of individual cells. These methods are crucial for understanding the causes of population effects in large-scale bioprocesses and for developing targeted control measures.

Bridging the monitoring gap between lab scales requires a “beginning-with-the-end-in-mind” approach in bioprocess development, combining high-resolution single-cell data with scalable inline sensors and multi-step bioprocess modelling. This enables the prediction of scale-dependent cell behaviour and opens up promising prospects for improving the scalability and robustness of industrial bioprocesses.

Keywords: Bioprocess monitoring, single-cell analysis, population heterogeneity, scale-up, robustness

T4.2 : Droplet-based microfluidics and automation to overcome challenges in the screening of secondary metabolites during bioprocessing

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Abstract :

Droplet-based microfluidics (DBM) technology and automated micro-scale miniaturization have developed considerable popularity for the analysis of heterologous single-cell growth as well as for the screening of secondary metabolites. Furthermore, DBM platforms are capable of producing a large number of samples within relatively short time frames. Given the enormous number of samples currently available, analytical methods compatible with this technology are required. However, there are limitations that create a bottleneck in their processing. These challenges are primarily related to volume availability. This study critically examines potential solutions to this challenge as well as the efficient integration of high-throughput (HTP) liquid handling platforms at scale-down level (MTP) with DBM platforms. Therefore, to rapidly process samples from DBM cultures, automated microlitre-scale liquid handling can facilitate the transfer to subsequent analytical stages. Furthermore, this HTP technology can be compatible with standard bioprocessing techniques designed for bacteria and cells, including microalgae. The integration of methods such as HPLC or SERS-Raman presents significant challenges for the practical use of DBM.

Keywords : Bioprocess, DBM, HTP, Automation

References :

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T4.3 : Model-based process design for surfactin production with *Bacillus subtilis*

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Abstract :

Bacillus subtilis is a key production organism in industrial biotechnology, but knowledge about fed-batch kinetics and time-dependent biological performance indicators like yields, growth rates and productivities remains limited, particularly regarding their variation over time during cultivation. Understanding these dynamics is essential for optimizing productivity, especially in the production of the promising biosurfactant surfactin. A kinetic model based on ordinary differential equations was developed to describe biomass growth, substrate consumption, surfactin synthesis and acetate formation in fed-batch cultivations of *Bacillus subtilis*. Based on data from 12 bioreactor experiments, the model proved to be robust and predictive across various feeding strategies. The kinetic model was used for a process design, in which the feeding phase was initiated directly from the beginning. In this process, an increase of the product titer of up to 46.33 g/L surfactin and a space-time-yield of 2.11 g/(L*h) was achieved. The kinetic model developed in this study enables the description of the time course of biomass growth, substrate consumption and product formation and thus significantly improves process understanding. The computation of key process parameters, which are not directly analytically accessible at any time, could thus be realized. In future work, the model will be extended to include in-situ product removal by foam fractionation, enabling a coupled description of fed-batch cultivation and surfactin separation. This integrated modeling framework will allow dynamic control of feeding and aeration based foam efflux, with the aim of achieving quasi-steady-state operation.

Keywords : *Bacillus subtilis*, Surfactin, Kinetic Modelling, Bioreactor

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Hiller, E., Off, M., Hermann, A., Vahidinasab, M., Benatto Perino, E.H., Lilge, L., Hausmann, R. (2024). The influence of growth rate-controlling feeding strategy on the surfactin production in *Bacillus subtilis* bioreactor processes. *Microbial Cell Factories*, *23*, 260. doi.org/10.1186/s12934-024-02531

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T 4.4 : Digital Twins in Modern Fermentation - Integration of high-predictive models in Autonomous Fermentation

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Abstract:

Despite advances in bioprocess engineering, fermentation development remains limited by biological complexity, non-linear system behaviour, and the experimental burden of exploring large design spaces. Traditional empirical optimisation often fails to capture the interplay between metabolism, regulation, and process conditions, particularly during scale-up.

Mechanistic modelling of the cell reaches a high accuracy in predicting complex behaviour of living organisms. Indeed, high-quality models can enable simulation of cellular responses across genetic or metabolic regulation and under environmental perturbation. Mathematical models transform fermentation R&D from trial-and-error to predictive, knowledge-driven workflows. Integration of this technology in digital twin frameworks where online measurements are analyses on-the-fly during the fermentation unlocks capability to control autonomously the bioreactor to avoid performance losses.

Coupling digital twins with laboratory automation and high-throughput experimentation enables iterative optimisation. iMEAN and AlgoTech join forces to embed iMEAN's high-predictive models into Bio-OS, AlgoTech's AI-native laboratory operating system, and its connected, self-piloted, autonomous bioreactor platform. The solution integrates mechanistic process simulation into real-time control, enabling deviation detection, adaptive strategies, and autonomous process management. Altogether, this work demonstrates readiness of digital twins' technology to accelerating development of scalable, industrially relevant, fermentation processes.

Keywords : Digital Twins, Fermentation Optimisation, Mechanistic modelling, Autonomous Industrial Bioprocess, AI-Driven Biomanufacturing

T 5.1: From Fundamental research regarding the protein N-glycosylation pathways in microalgae to the production of microalgae-made biologics at industrial scale

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Abstract:

After screening different microalgae from their capacity to synthesize glycoproteins, we identified that the diatom *Phaeodactylum tricornutum* was able to synthesise hyman type N-glycans to its endogenous proteins. Therefore, we decided to demonstrate that is would be possible to use such microalgae as an alternative cell biofactory for the production of antibodies. As compared to mammalian cells as an expression system, the use of microalgae helps us to reduce the production costs of the antibodies while improving their quality and efficacy, thereby facilitating patient access to these innovative treatments. Moreover, this represents a sustainable way to produce antibodies as demonstrated through the production of glycosylated functional antibodies directed against viruses like Hepatitis B and Marburg viruses (Hempel et al., 2011; Hempel and Maier, 2012; Vanier et al., 2015; Hempel et al., 2017; Vanier et al., 2018).
In this context, we created in November 2021, ALGA BIOLOGICS, a deeptech company that is producing antibodies for diagnostic and therapeutic purpose in microalgae like *Phaeodactylum tricornutum*. To achieve its first proof-of-concept at 200L scale, ALGA BIOLOGICS is currently producing an antibody directed against neuroblastoma, a pediatric cancer that affects approximately 25,000 young children worldwide. In this presentation, in vitro functional characterization of this microalgae-made monoclonal antibody will be presented. The company is supported in its ambition by the French government through the France 2030 program under the acceleration strategy for bioproduction and biotherapies and by the ADD (Aide au Développement Deeptech) from BPI France.

Keywords: Microalgae, Bioproduction, Antibodies, Biologics, Sustainability

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T 5.2 : From ideation to execution: key stages for industrial bioprocess development

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Abstract:

Advances in life science research technologies continually enhance the understanding of biological systems, thereby fostering innovation in biotechnology and making microbial fermentations increasingly significant in modern economies. However, the successful commercialization of novel biobased products requires more than laboratory-scale prototyping; it necessitates comprehensive evaluation criteria encompassing the entire process and value chain, which demand considerable time and investment. This presentation will examine challenges associated with the innovation ambition matrix in industrial biotech companies and explore various factors involved in progressing from ideation to commercialization of industrial biobased products—elements frequently underestimated in academic settings and by start-up ventures.

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Keywords : Industrial scale-up, Bioprocess optimization, Techno-economic performance

T.5.3 : Microphyt-From Lab to Market: Scaling Microalgae Production and Extracts for Industry

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Abstract:

Microphyt's journey began with a vision: unlocking the potential of microalgae to create high-value ingredients for cosmetics and nutritional supplements. Microphyt has mastered the entire value chain, from strain selection to large-scale production, ensuring quality and performance.
A key milestone was scaling up production using advanced photobioreactors, which allow precise control over growth conditions. This expertise sets Microphyt apart, enabling the production of premium microalgae-based ingredients with consistency and efficiency. The company now operates a state-of-the-art facility with 315 m^3 of culture volume and an Atex-certified extraction workshop.
Microphyt also handles every aspect of the process—from R&D and production to regulatory and quality control, and marketing. Furthermore, Microphyt is developing a strategy to validate these microalgae-derived ingredients, all the way through to clinical trials in humans for use in cosmetics and dietary supplements.
While Microphyt has achieved significant progress, new challenges remain, such as expanding applications. Our story is one of innovation, precision, and a commitment to delivering microalgae-based solutions to the market.

Keywords: Microalgae, Industrialization, Extraction, Cosmetic, Food supplement.

T.5.4 : From Innovation to Production: Mastering the Art of Industrial Scale-up to Secure Your Path to Market

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Abstract :

Industrial scale-up is a critical step in transforming biotechnological innovation into viable commercial production. ARD leverages over 30 years of expertise and integrated infrastructures (from laboratory to pilot and demonstration scales) to support this transition. The bioprocess value chain includes biomass production, fermentation or biocatalysis, and downstream processing (DSP), each playing a key role in overall performance.

Process success is primarily driven by microorganism performance (titer, yield, productivity) and by the optimization of both upstream (fermentation) and downstream operations. High titers simplify DSP, high yields reduce operating costs, and high productivity lowers capital investment requirements. However, increasing product purity significantly raises process complexity and cost.

Scaling up from lab to industrial level introduces major challenges, including oxygen transfer, mixing, variability in operating conditions, and maintaining reproducibility. Anticipating these effects through downscaling approaches and adapting process recipes is essential to ensure robust performance at scale.

A structured scale-up methodology is required, progressing from feasibility studies to lab development, pilot trials, industrial demonstration, and finally production. This approach integrates technology transfer, process adjustments, and performance validation, including mass balance and compliance with product specifications.

Ultimately, successful scale-up delivers proven industrial feasibility, reliable techno-economic data, and initial production batches, while significantly reducing risks. Leveraging specialized platforms accelerates time-to-market, minimizes investment, and secures a more reliable path from innovation to full-scale production.

Keywords : Industrial scale-up, Bioprocess optimization, Techno-economic performance

T 5.5 : Industrial deployment of recycled-carbon fermentation: A translational framework for low-emission bioprocesses

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Abstract:

This works focuses on the industrial deployment of recycled-carbon fermentation for developing low-emission bioprocesses. The approach aims to reduce greenhouse gas emissions while supporting the transition from refined sugar-based feedstocks toward circular and recycled carbon sources. It emphasizes the valorisation of industrial side streams, such as whey-derived substrates and other carbon rich coproducts, as sustainable fermentation inputs. Regulatory compliance must be embedded from the design phase to guarantee consistent product specifications, especially for food and bio-based applications. Beyond technical considerations, recycled-carbon fermentation aligns with European sustainability policies and strengthens Scope 3 emission reduction strategies. Measurable carbon footprint reduction enhances sustainability reporting, improves eligibility for green finance, and reduces transition risk. Although proof of concept studies has demonstrated technical feasibility, the main challenge lies in translating these concepts into robust, stable, and economically viable large-scale operations. Successful implementation requires managing substrate variability, ensuring fermentation stability, maintaining downstream processing compatibility, and controlling contamination risks. Galactic has explored several low carbon emission raw materials for fermentation and has evaluated them based on several factors such as raw material carbon concentration, accessibility and stability, impact on the fermentation kinetics as well as the ease of applying (traditional) post-treatment methods. This study provides industrial partners with additional selection criteria, beyond the cost of the raw material and the potential savings in carbon emission, when commencing projects on low-emission bio-processes.

Keywords: Recycled-carbon fermentation, carbon footprint reduction

T 5.6 : Adoption of miniaturized an automated approaches to assist R&D for bioprocess developments

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Abstract :

The growing adoption of miniaturization and high-throughput technologies is transforming microbial biotechnology by enabling faster, data-rich experimental cycles. At TWB, a biofoundry dedicated to microbial process innovation, these approaches are used to accelerate strain-engineering, screening, and bioprocess development. Automated platforms allow the parallel execution of thousands of experimental events with improved reproducibility and traceability, expanding the accessible biodiversity of engineered and natural microbial strains. Miniaturized fermentation systems, ranging from microscale bioreactors to droplet- or plate-based cultures, facilitate the exploration of vast design spaces while minimizing reagent costs and material waste. They enable cost-effective hypothesis testing and rapid process optimization. However, the reduction in working volumes introduces challenges in scaling, process control, and representativeness of larger-scale conditions. To address these limitations, TWB integrates modeling and algorithmic approaches—including data-driven design and machine learning—to strategically reduce the experimental search space and guide experimental decisions. These computational tools, developed through collaborations and consolidated within TWB ecosystem, help bridge the gap between microscale findings and scalable processes. The communication will present examples and results illustrating both the benefits and constraints of miniaturized, high-throughput experimentation in advancing microbial biotechnology within a biofoundry framework.

Keywords: Biofoundry ; HT experiment ; Process optimisation ; Upscale ; Downscaling

T 5.7 : Bioindustry 4.0 : Leveraging advanced digital solutions to empower the European bioindustry sector

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Abstract :

Industrial biotechnology, considered a cornerstone of the circular economy, is data-intensive and an ideal candidate for the use of advanced digital methods. It is also a challenge on many levels, including the unpredictability of live matter.

Spearheaded by six major European research infrastructures and supported by a consortium of 25 organizations, Bioindustry 4.0 mission is to enhance the competitiveness of the European biotech sector globally.

Leveraging advanced digital solutions – including next generation strain searches, AI powered digital twins, and new sensors – will help us to create and optimise innovative and reliable bioprocesses. These services are being developed for European research infrastructures, which will benefit researchers from industry and academia alike.

Services developed by partners engaged in Bioindustry 4.0 are expected to be unavoidable tools to support bioprocess from lab to industrial scale.

This project is funded by the European Union under Grant Agreement n° 101094287.

Keywords : Digital twin, data fabric, smart biomanufacturing, research infrastructure

References :

Bioindustry 4.0, European Union under Grant Agreement n° 101094287, Cordis 10.3030/101094287

Poster presentation

P 1.1 :Lactic acid fermentation enhances antioxidant potential of chicory by-products for functional food applications

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Abstract:

Chicory (Cichorium intybus) processing generates substantial volumes of unused by-products such as peelings and roots, rich in polyphenols and bioactive compounds with potential health benefits. This study investigated lactic acid fermentation as a sustainable strategy to valorize these residues into functional food ingredients. Eight strains of *Lactiplantibacillus plantarum* and *Lactobacillus reuteri*, including two isolates from the native microbiota of Witloof chicory (Belgian endive), were tested for their ability to ferment four by-product types. Belgian endive peelings (EPBU) proved the most promising substrate based on bacterial growth and antioxidant activities. Antioxidant capacity was assessed using a DPPH assay and a cellular ROS assay in hepatic and intestinal cell lines. Fermentation with strain 31c significantly enhanced ROS scavenging, achieving up to a 17-fold reduction, among the strongest effects in fermented plant matrices. Metabolomic analysis of fermented EPBU revealed chlorogenic acid and increased compounds linked to fermentation pathways. These results demonstrate lactic acid fermentation as an effective strategy to improve chicory by-product bioactivity for sustainable food and nutraceutical applications.

Keywords:
Chicory by-products; Lactic acid fermentation; Antioxidant activity; *Lactiplantibacillus plantarum*;

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P 1.2: Screening of Yeast Based Nutrients to optimize growth of Lactic Acid Bacteria at 96-well MTP scale and validation at AmbR250 â scale

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Abstract :

Procelys by Lesaffre is a key manufacturer of Yeast-based Nutrients (YBN), which are used in the probiotic industry to meet the diverse nutritional requirements of probiotic strains and optimize their growth. This study outlines the development of a semi-automated high throughput screening strategy at 96-well microtiter plate (MTP) scale to identify optimal YBN for the production of probiotic lactic acid bacteria (LAB).

A total of 118 novel YBN prototypes with diverse compositions were produced at lab scale and screened at 96-well microtiter plate scale by assessing their ability to support the growth of 27 lactic acid bacterial strains across 9 different species. Performance of YBN was evaluated by monitoring growth kinetics (OD_{600nm}) over 48h, with prototypes ranked according to maximum growth rate and maximum optical density reached.

This screening approach identified the five most promising YBN prototypes, which supported comparable or superior growth relative to a reference condition for most LAB screened. The production of these five YBN nutrients was scaled up, and evaluated in AmbR250â bioreactors. The results demonstrated growth that was generally similar to or better than the reference condition, and the performance ranking remained consistent with the initial screening, confirming the reliability of the 96-well MTP screening method and analysis workflow set-up.

Furthermore, while the screening identified YBNs that were broadly effective across most LAB species tested, performance remained strain-specific. Growth could be further optimized through strain-specific analysis and tailored nutritional solutions.

Keywords: Yeast Based Nutrients; Lactic acid bacteria; MTP screening

P 1.3: Bioproduction and sustainable supply of the anticancer compound etoposide by yeast cell-factories

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Abstract:

Lignans are a family of specialized metabolites produced by numerous plants with broad pharmaceutical applications. Among them, etoposide is an anticancer compound produced by semisynthesis from podophyllotoxin extracted directly from *Podophyllum hexandrum*. This production method places significant pressure on the plant, which is difficult to cultivate and increasingly affected by global warming, leading to significant fluctuations in supply. Moreover, as observed during the COVID pandemic, the scarcity of these raw materials makes importation much more difficult.

To ensure a sustainable supply, relocating production by developing new strategies is essential to ensure a sustainable access to this compound for patients. Among these strategies, the bioproduction of this metabolite, achieved using yeast engineered with heterologous plant genes, offers the advantage of enabling more stable and consistent local production. This eliminates dependence on original plant resources, particularly when combined with a systemic bioprocess engineering approach.

In this context, our work aimed to refine the design and construction of a *Saccharomyces cerevisiae* strain expressing plant-derived genes ensuring the production of 4'-demethylpipodophyllotoxin (4'dEPT), a new precursor, enabling etoposide production through fewer semisynthetic steps. We subsequently implemented a rational, controlled cultivation process, combining optimised feeding strategies with suitable physicochemical parameters. This 4'dEPT-producing strain enabled us to achieve, through bioconversion, production of 553 mg. L-1 of 4'dEPT, representing the highest concentration ever reported to date across all production models.

The next step will be to further optimize upstream process steps to achieve economically viable levels for industrial-scale use.

Keywords: 4'-Demethylepipodophyllotoxin, Bioprocess, Bioconversion, Scale-up, cell factory

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P 1.4: Medium optimisation using *Yarrowia lipolytica* for recombinant food protein production, improving economic feasibility at industrial scale.

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Abstract:

Global population growth results in a significant increase in demand for dietary protein and the current production methods (agriculture and breeding) will not be able to sustainably supply the projected global population[i]. Precision fermentation offers an innovative solution by employing engineered micro-organisms to produce animal proteins of interest for food applications[ii]. Still a challenge remains for production at affordable costs. Many factors can have an economic impact such as the selected microorganism, the medium composition or the purification method[iii]. We focused our study on the yeast *Yarrowia lipolytica* (YL), for its ability to grow on many substrates and even agricultural wastes. Furthermore, its GRAS status is accordance with the european Novel Food regulation. We transformed a YL Po1g strain with a recombinant plasmid integrating a secretion signal, to produce a Recombinant Food Protein (RFP) (confidential). Strains were selected with the Yeastern Blot immunostaining method[iv]. Protein production levels was assessed with ELISA method, on different defined media, at a flask scale. The most promising medium was then optimised using a Plakett burman experimental design for the assessment of the main composants and the carbon and nitrogen sources[v]. These optimisations allowed the reduction of the medium cost by 75% (protein production yield is still to be assessed).

Keywords: fermentation, yeast, food protein, medium optimisation

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P 1.5: Enhancing probiotic bacteria growth via balanced RNA and DNA precursors in an innovative fermentation nutrient

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Abstract :

Probiotic bacteria have gained significant attention due to their proven benefits for human health. Among the most common are species such as *Lactobacillus*, *Bifidobacterium*, and *Bacillus* [1]. Their growth and viability are highly dependent on environmental and nutritional conditions, making it essential to understand their specific requirements [2-4]. In this context, Lallemand Bio-Ingredients and AS TFTAK investigated the role of nucleotides, nucleosides, and nucleobases in supporting probiotic lactobacilli growth.

Three strains (*Lactobacillus rhamnosus*, *Pediococcus acidilactici*, and *Lactobacillus acidophilus*) were cultivated in 2L bioreactors using media containing different yeast-derived nitrogen sources: one rich in nucleotides, one containing nucleosides and nucleobases, and a standard yeast extract.

Results showed variability in the ability of these bacteria to synthesize nucleotides. Some strains exhibited auxotrophy and relied on salvage pathways, requiring external nucleobases or nucleosides. Notably, a balanced combination of RNA and DNA precursors significantly improved *L. rhamnosus* growth. The yeast extract containing nucleosides and nucleobases reduced the lag phase by 50% and increased viable cell counts sixfold.

These findings demonstrate the importance of tailored nutrient formulations and highlight the benefit of combining complementary yeast-derived ingredients.This approach could be extended to more demanding microorganisms, such as *Bifidobacterium* and microbiome bacteria.

Keywords: Nucleotides, Nucleosides, Nucleobases, Yeast based nutrient

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P 1.6: High-Throughput Screening of Promoters for Enhanced Cyclic Lipopeptide heterologous Production in Pseudomonas

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Abstract:

Cyclic lipopeptides (CLiPs) from Pseudomonas are promising antimicrobial compounds for environmental and industrial biotechnology. However, their exploitation remains limited by low yields, high production costs, and complex co-production profiles.

To overcome these limitations, the HETEROCLIPS (ANR-22-CE20-0044) project combines heterologous expression and process engineering to enhance targeted CLiPs production. This approach needs the selection of the most active promoters to drive lipopeptide biosynthetic clusters relying on transcriptional reporter systems where GFP fluorescence indicates promoter strength. Accordingly, five promoter constructs were screened in Pseudomonas fragi FL4BN2 using high-throughput BioLector micro-fermentations. Real time monitoring over 120 h successfully identified promoter PnadC as the strongest candidate, followed by promoter Ptac, both exhibiting a significant increase in expression activity compared to the parental strain. Conversely, promoters Picd_2, PrpsA_2, and Psyp showed baseline fluorescence levels similar to the wild type control. Overall, these findings pave the way for integrating PnadC and Ptac upstream of CLiPs biosynthetic gene clusters to achieve scale-up of these potent microbial biopesticides.

Keywords: Cyclic lipopeptides, Pseudomonas, High-throughput promoter screening, Micro-fermentations, Biopesticides.

P 1.7 : Study of the factors influencing the robustness of genetically modified strains of *C. necator* for the production of hydrocarbons from CO2 in a bioreactor

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Abstract :

Growing concerns over fossil resource depletion have increased interest in microbial biofuel production from renewable carbon sources. Alkanes and alkenes, the main components of petrol, diesel and kerosene, can be synthesized through engineered fatty acid–derived pathways thanks to microbial biosynthetic enzymes (Peralta-Yahya and Keasling, 2010). The bacterium *Cupriavidus necator* is a promising candidate for biofuel production from CO_2. However, producing these non-natural molecules imposes metabolic burden and toxicity on host cells, leading to phenotypic heterogeneity, process instability and reduced yields. In this study, *C. necator* was engineered to produce alka(e)nes using plasmids. Plasmids carry *Synechococcus elongatus* genes involved in fatty acid conversion. Cultivations carried out in gas-bottle flasks with a 1 L headspace containing a defined mixture of 8% O_2, 74% H_2, and 18% CO_2 confirmed both autotrophic growth and alka(e)nes production after arabinose induction under decane extractive fermentation conditions. The most productive strain was then cultivated in a 1 L gas-fed bioreactor, achieving the highest reported alka(e)ne yield in bibliography for *C. necator* under autotrophic conditions (Crépin *et al.*, 2018). To investigate phenotypic heterogeneity and plasmid instability, plasmid presence was monitored by a plasmid-encoded fluorescent reporter (eGFP) detected by flow cytometry, and copy number was quantified by digital PCR.

Keywords : *Cupriavidus necator*, Autotrophy, Gas cultivation, Bioprocesses, Heterogeneity.

Références :

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Peralta-Yahya, P.P., and Keasling, J.D. (2010) Advanced biofuel production in microbes. *Biotechnol J* **5**: 147–162.

P 1.8: Metabolic response of *Lactiplantibacillus plantarum* to compounds of *Himanthalia elongata*: an approach to unlock lactic acid bioprocessing of brown seaweeds.

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Abstract:

Lactic acid fermentation represents an innovative and sustainable approach for the bioprocessing of natural biomass. Aqueous extracts of the brown seaweed *Himanthalia elongata*, which is widespread along the Atlantic coast, exhibit promising antioxidant and anti-inflammatory properties [1]. However, the structural complexity of its cell wall and its rich biochemical composition cause significant technical challenges for lactic acid fermentation.

The fermentability of seaweed-derived compounds, as well as their potential inhibitory effects on lactic acid bacteria, were evaluated using a modified De Man–Rogosa–Sharpe medium. Key parameters of fermentation process - including temperature, inoculum size, pH, aeration, carbon source (mannitol, glucose, galactose and alginates), and inhibitory compounds (sodium chloride, potassium chloride and phloroglucinol)—were optimized. The metabolism of *L.plantarum* was monitored through the quantification of organic acids and monosaccharides using chromatographic methods, alongside measurements of pH and bacterial growth. *L. plantarum* showed concentration-dependent responses to salts and phloroglucinol. NaCl and KCl delayed acidification, growth, and lactic acid production at high concentrations, although pH still decreased below 4,5. KCl was less inhibitory than NaCl. Phloroglucinol strongly impaired bacterial activity, completely preventing acidification and lactic acid production at 10 g/L after 48h of fermentation. Galactose and mannitol supported rapid growth and acidification, whereas fucose and alginate allowed growth without acid production. A carbon mixture representative of *Himanthalia elongata* composition supported efficient growth, acidification, and preferential utilization of glucose and galactose.

Further investigations using whole seaweed extracts are required to confirm the feasibility and efficiency of *H.elongata* fermentation.

Keywords: *Himanthalia elongata;* Lactic acid fermentation; Carbon Metabolism; Kinetics; Bioprocessing

References:

Castetbon C, Jabbour C, Burlot A-S, Le Penven S and Bedoux G (2026) The potential applicability of Himanthalia elongata fermentation for the production of nutraceuticals. Front. Mar. Sci. 13:1741035. doi: 10.3389/fmars.2026.1741035

P 1.9: Bacterial Strain Engineering to Enhance the Heterologous Production of the Cyclic Lipopeptide Orfamide

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Abstract:

Cyclic lipopeptides (CLiPs) are microbial secondary metabolites with diverse functional properties and biological activities, making them promising biocontrol agents. However, their natural production is limited and often results in complex mixtures, restricting their characterization and practical applications. Heterologous production represents an attractive strategy to overcome these limitations by expressing lipopeptide synthetase gene clusters (BGCs) in a suitable non-native host organism. In this study, we focused on optimizing the heterologous production of orfamide, a cyclic lipopeptide synthesized by *Pseudomonas* spp. We demonstrated that orfamide is likely non-toxic to *Paraburkholderia phytofirmans* PsJN, supporting its suitability as a heterologous host. Reverse Transcription-Polymerase Chain Reaction (RT-PCR) analyses confirmed the co-transcription of the orfamide synthetase-encoding genes, while promoter engineering increased their transcription levels. In addition, Transformation-Associated Recombination (TAR) cloning enabled the successful assembly and insertion of these genes into expression vectors for future integration into the genome of *P. phytofirmans* PsJN for heterologous orfamide production. These results provide a strong basis for developing alternative heterologous systems for CLiPs production. To further support this objective, we performed large-scale genome mining analyses of *Pseudomonas* genomes and identified strains with high CLiP biosynthetic potential, representing valuable reservoirs of biosynthetic pathways for future heterologous production strategies, as well as strains producing none or only a single lipopeptide, constituting promising chassis for heterologous expression approaches.

Keywords: cyclic lipopeptides, biocontrol, *Pseudomonas*, transformation-associated recombination cloning, heterologous production

P 1.10: Scale-up of a fermentation process for the production of *Pseudomonas* active lipopeptides

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Abstract :

The genus *Pseudomonas* is capable of producing lipopeptides, which are secondary metabolites. These possess antimicrobial properties, amongst others. In *Pseudomonas syringae*, three families have been identified: factins, mycins and peptins (Bricout et al., 2022). The aim of this study is to improve the production of *P. syringae*'s lipopeptides in a bioreactor to exploit the full potential of their properties.

The impact of culture conditions on lipopeptide production was assessed in 500 mL flask. The most favourable conditions were then selected for scale-up in a 5 L stirred tank reactor (STR) using modified SRM (Syringomycin Recovery Medium) (Gross, 1985). Lipopeptide production is quantified by HPLC-MS.

The culture in a STR promoted the growth of P. syringae. The maximum biomass was increased by 50% and the growth rate (μmax) rised from 0,11 to 0,15 h^{-1}. Likely due improved oxygenation, as the theoretical oxygen mass transfer coefficient (kL.a) is increased from 24.5 h^{-1} to 272.5 h^{-1}. However, this did not allow to increase the overall lipopeptide production. Differences in lipopeptide production highlight the challenges of scaling up from 500 mL flask to 5 L STR. Optimising operating parameters, such as aeration or fermentation mode (batch, fed-batch, or continuous), is required to enhance lipopeptide production, currently under investigation.

Keywords: Fermentation, bioreactor, lipopeptides, P. syringae, biocontrol

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Bricout, A., Morris, C.E., Chandeysson, C., Duban, M., Boistel, C., Chataigné, G., Lecouturier, D., Jacques, P., Leclère, V., Rochex, A., 2022. The Diversity of Lipopeptides in the Pseudomonas syringae Complex Parallels Phylogeny and Sheds Light on Structural Diversification during Evolutionary History. Microbiol Spectr 10, e0145622. https://doi.org/10.1128/spectrum.01456-22

Gross, D. c., 1985. Regulation of syringomycin synthesis in Pseudomonas syringae pv. syringae and defined conditions for its production. Journal of Applied Bacteriology 58, 167–174. https://doi.org/10.1111/j.1365-2672.1985.tb01444.x

P 1.11: Enhanced fengycin production in a non-GMO *Bacillus subtilis* strain via culture and medium optimization

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Abstract:

Bacillus species are well-known producers of secondary metabolites, particularly lipopeptides which are remarkable antimicrobial compounds. *Bacillus subtilis* produces three major lipopeptide families, including fengycin, a highly effective antifungal lipopeptide. The aim of this work is to optimise fengycin production from a non-GMO fengycin monoproducer mutant (derivates from the strain ATCC 21332) by screening culture conditions and medium composition.

For culture conditions optimisation, several parameters were evaluated in flasks using Landy medium, including temperature (23–37 °C), oxygen transfer expressed as the volumetric mass transfer coefficient k_L a (9.23, 16.41, and 29.17 h^{-1}), and initial inoculum size (OD600 0.1– 0.5). For medium optimisation, Landy medium was used as a reference, and carbon and amino acid sources were varied. The final experiments were performed in microplates using a BioLector system, fengycin production was quantified by UHPLC and HPLC-MS.

These experiments resulted in a 39% increase in fengycin production by increasing the inoculum size and incubating at 30 °C in flasks, and up to a 68% increase under optimised oxygen transfer conditions. Medium optimisation further resulted in a 200% increase in production. The combination of optimised conditions will be used to upscale fengycin production from this non-GMO strain at semi-pilot scale, followed by in vivo evaluation on host plants within the Interreg Biocontrol 4.0 project portfolio.

Keywords: biocontrol, lipopeptides, Bacillus subtilis, Fengycin, optimisation

P 1.12: Generation of *Bacillus subtilis* non-GMO monoproducers and overproducers of surfactin and fengycin by UV-induced random mutagenesis

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Abstract:

Bacillus subtilis is a natural producer of lipopeptides, including surfactin and fengycin, which show potent antimicrobial and antifungal activity even at low concentrations. These compounds are promising bio-based alternatives for the control of plant diseases such as apple scab, a major issue in Wallonian organic fruit production, where copper and sulfur-based treatments currently prevail. The urgency to shift towards these biomolecules is driven by two converging challenges: the progressive restriction of copper use by the European Commission, now officially classified as a candidate for substitution, and the naturally low lipopeptide yields of wild-type strains, which remain a key bottleneck for large-scale production, compounded by the strict prohibition of GMO strains in organic farming. To address these challenges, we developed a UV-induced random mutagenesis strategy and generated three non-GMO mutant strains derived from *B. subtilis* ATCC 21332, each suited for different applications. The first mutant co-overproduces both surfactin and fengycin (2.32-fold and 2.07-fold over the wild-type, respectively). The two additional mutants are monoproducers, each exclusively producing either surfactin or fengycin. Notably, the fengycin monoproducer also displays a 2-fold increase over the parental strain. These monoproducer strains are expected to greatly simplify downstream purification and lower production costs by eliminating the need to separate co-produced lipopeptides.

Keywords: biocontrol, lipopeptides, *Bacillus subtilis*, non-GMO, overproduction

P 1.13: Cell viability assessment post blue light exposure for optogenetic strain use in bioproduction

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Abstract:

This study addresses the heterogeneity of cell populations within an axenic culture in a fermenter. These subpopulations directly impact the performance and reproducibility of the process. The objective of this study is to monitor the physiological states of cells during culture using online measurements and to manipulate fermenter parameters via a feedback loop to guide cells along the desired pathway and limit subpopulations.

For this study optogenetic strains of *Saccharomyces cerevisiae* (which produce a compound of interest in response to light) are used. These strains produce GFP when exposed to blue light. Cell monitoring is performed by flow cytometry with and without labeling. The markers used are indicators of membrane permeability as well as the production of reactive oxygen species (ROS). Cultivability is measured by plating and compared with flow cytometry.

Initial cultures highlighted the harmful effects of blue light on yeast. Upon blue light exposure, an increase in ROS levels and membrane permeability was observed, indicating impaired cell viability. The impact of light on viability and cultivability was subsequently evaluated at three different intensities compared to a control culture. These measurements allow for the determination of the optimal light intensity range to limit the phototoxic effect.

Keywords: Yeast – Optogenetic – Bioproduction - Cytometry

P 1.14: Recombinant therapeutic mRNA production in yeast: strategies to increase clone productivity

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Abstract:

The Covid-19 pandemic highlighted mRNA as a promising therapeutic modality, not only for vaccines but also for oncology and rare diseases. Today, therapeutic mRNA is produced via in vitro transcription (IVT), a rapid and safe method that enzymatically synthesize mRNA from a DNA template. However, its limitation to simple, small mRNA and high manufacturing cost has spurred interest in recombinant mRNA production as an alternative. bYoRNA's patented technology leverages recombinant yeast cells to produce therapeutic mRNA via fermentation - a novel, disruptive approach offering safe and affordable supply. However, a key challenge remains: developing high-producing, stable yeast strains suitable for pharmaceutical production.

To tackle this challenge, several strategies were explored to enhance clone productivity, and maintain stability over generations: (i) optimisation of the expression vector through the addition of strong promoters; and (ii) optimisation of the clone selection process through the use of a novel selection marker and optimized selection conditions.

These combined approaches successfully increased clone productivity to a level compatible with our target, a critical step toward fermentation and purification process industrialization and efficient mRNA production for pharmaceutical use.

Keywords: Therapeutic mRNA; Strain engineering; Promoters; Selection marker

P 2.1: Bioproduction and modulation of the composition of Single Cell Protein (SCP)

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Abstract:

In the context of climate change and growing global population, alternative sources of protein are needed to ensure global food security. Among the proposed solutions, microbial biomass—commonly referred to as Single Cell Protein (SCP)—stands out due to the rapid growth rate of microorganisms, their high protein content and the fact that their production does not depend on seasonal variations. The project µO-PROT (PIA-France2030/ANR-23-DIVP-0001), through an integrated multidisciplinary approach combining bioproduction, downstream processing, characterization of biochemical, functional, toxicological and nutritional properties, as well as societal issues will assess the potential of crude- and processed- SCPs as an alternative protein source for human nutrition.

Within this context, the objective of this work is to explore different fermentation strategies to modulate the composition of SCP, particularly their protein, lipid, nucleic acid, and carotenoid content. To this end, the use of various yeasts and bacteria, such as *Y. lipolytica*, *R. glutinis*, or *C. necator*, a bacterium capable of utilizing CO_2, was performed under different culture conditions. For example, it has been shown that controlling the growth rate in a continuous process allows for the modulation of nucleic acids content, compounds that can cause health problems such as gout. Furthermore, controlling the N/C ratio through specific feeding strategies affects lipid, protein and nucleic acid content.

Keywords: Single Cell Protein (SCP), Bioproduction, Yeast, Bacteria, Food

P 2.2: *Lactobacilli* consortium and *Corynebacterium glutamicum* sequential cultivation to produce single cell protein (SCP) from seaweed by-products

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Abstract:

The demand for sustainable protein sources is steadily increasing. Single cell protein (SCP) provides amino acid profiles suitable for human nutrition, but high production costs remain a major bottleneck. Although various industrial by-products have been explored as SCP growth substrates, nutrient-rich residues from the marine industry remain underutilized, representing an opportunity to enhance the sustainability and cost-effectiveness of SCP production. After agar removal, the macroalga *Gelidium corneum*, a widely used species in the agar extraction industry, retains carbohydrate and protein fractions with interesting nutritional profiles, which are not readily bioaccessible. Saccharification of the algal crude fiber followed by microbial fermentation enhances protein bioaccessibility and quality and also provides improved bioactive properties. Industrial residues of *Gelidium corneum* were enzymatically hydrolyzed into a glucose-rich syrup and subsequently used as fermentation substrate with a consortium of four lactic acid bacteria (4LAB). Afterwards, the whole broth was separated into two fractions: protein-rich biomass pellet, and lactic acid-rich supernatant. The supernatant was further utilized to cultivate *Corynebacterium glutamicum* for additional SCP enrichment. Analyses revealed higher protein content in both 4LAB pellet (37.4 ± 0.7 g/100g DW) and whole *C. glutamicum broth* (32.8 ± 0.8 g/100g DW) when compared with residual alga (18.5 ± 0.05 g/100g DW); all fermented products exhibited higher protein bioaccessibility and protein quality than original residues. *C. glutamicum product* displayed stronger antioxidant properties than 4LAB fermented biomass. These findings demonstrate the potential of upgrading marine by-products with sequential microbial fermentations to produce nutritional and functional SCP-enriched ingredients for food or feed applications.

Keywords: Macroalga, Sequential fermentation, Single cell protein, Lactic acid fermentation, *Corynebacterium glutamicum*.

P 2.3: Tailored Bioplastics for Biomedical Applications: Controlling 4HB Incorporation in PHA Copolymers

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Abstract:

Polyhydroxyalkanoates (PHAs) are biodegradable polymers accumulated intracellularly by various microorganisms as carbon and energy reserves under nutrient-limiting conditions. Among them, polyhydroxybutyrate (PHB) has attracted considerable attention as a sustainable alternative to petroleum-derived plastics due to its biocompatibility and complete biodegradability. Nevertheless, the PHB homopolymer is limited in practical applicability because of its brittleness, high crystallinity, and narrow processing window, resulting from its rigid, highly ordered chain structure. To overcome these drawbacks, the incorporation of additional monomeric units into the polymer chain has been extensively investigated [1]. Increasing incorporation of 4-hydroxybutyrate (4HB) significantly enhances polymer ductility while influencing physicochemical properties such as crystallinity and surface morphology [2]. Higher 4HB fractions generally reduce the melting temperature, glass transition temperature, and tensile strength, while increasing elongation at break [3], promoting a transition from a rigid thermoplastic material to one with elastomeric characteristics. This broad tunability of material properties makes P(3HB-co-4HB) attractive for a wide range of applications [4]. Since 4HB occurs naturally in the human body and P(3HB-co-4HB) demonstrates excellent biocompatibility, P(4HB) is currently the only FDA-approved PHA for medical applications [5]. The combination of biodegradability and biocompatibility makes this copolymer promising for biomedical applications [3].

This work explores strategies to control the 4HB molar fraction in P(3HB-co-4HB) copolymers produced by *Cupriavidus necator* through manipulation of γ-butyrolactone feeding strategies and dissolved oxygen concentration to tailor polymer properties for biomedical applications.

Keywords: Polyhydroxyalkanoates (PHAs), P(3HB-co-4HB) copolymers, *Cupriavidus necator*

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P 2.4: Controlling microbial population instability in continuous cultivation: impact of switching cost and phenotypic escape

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Abstract:

Continuous cultivation offers a promising alternative to batch and fed-batch processes by enabling sustained production and reduced downtime. However, its broader application is limited by population instability, affecting both productivity and process robustness. While such instability is often attributed to genetic mutations, increasing evidence highlights the role of phenotypic heterogeneity within isogenic populations [1]. In this context, phenotypic escape emerges as a rapid and reversible process leading to subpopulations with reduced gene expression [2].

To investigate phenotypic escape, low and high switching cost systems were compared using *Escherichia coli* BL21 (low switching cost) and BL21(DE3) (high switching cost). Switching cost is defined as the reduction in growth rate associated with the activation of a target gene circuit. In *E. coli* BL21(DE3), lactose induces the T7 expression system, resulting in a significant growth penalty and thus a high switching cost. Preliminary experiments in microtiter plate cultures with glucose-lactose mixtures revealed classical diauxic growth with two exponential phases under low switching cost conditions, mainly due to adaptation from glucose to lactose. Interestingly, high switching cost conditions resulted in an unexpected third exponential phase, suggesting complex population dynamics. These results indicate that switching cost may promote phenotypic diversification and contribute to phenotypic escape.

Future work will combine lab-scale bioreactor cultures with automated flow cytometry to clarify the relationship between switching cost, phenotypic escape, and population instability. Ultimately, this work aims to provide new insights into controlling heterogeneity in continuous cultures, paving the way for more robust and predictable biomanufacturing systems.

Keywords: phenotypic escape, switching cost, phenotypic heterogeneity, continuous culture, *Escherichia coli*

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P 2.5: A ReacSight platform for characterizing genetic instability in synthetic biology

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Abstract:

Reprogramming living systems for bioproduction imposes a synthetic burden on host cells, triggering stress responses and growth defects. While research has primarily focused on understanding the primary burden effect—the direct impact of engineered systems on host-cell physiology and growth[1–3]—much less attention has been given to its long-term consequences on the genetic stability of bioproduction systems. By imposing a fitness cost, synthetic burden creates selective pressure for cells that lose or silence the engineered construct, enabling non-producing mutants to outcompete functional cells. Studying these evolutionary dynamics is experimentally challenging because it requires continuous induction and long-term single-cell monitoring. To address this, we extended our ReacSight strategy[4] into a platform built around commercial eVOLVER bioreactors (FynchBio), integrating flow cytometry and robotic liquid handling for up to 16 parallel long-term experiments with single-cell resolution and optogenetic induction. Validation in S. cerevisiae demonstrated stable turbidostat operation at OD 0.5 and 30°C over 2-week runs with automated cytometry every 12–24 hours collecting 5,000 events per reactor. Through optogenetic pulse-width modulation, the platform enables precise gene expression control, linear with duty cycle (R^2=1.00). With long-term automated operation, tunable optogenetic induction, and single-cell resolution, ReacSight directly overcomes the barriers to characterizing genetic instability, while its commercial hardware and open-source software make it broadly accessible across synthetic biology applications.

Keywords: synthetic biology, automation, optogenetics, bioproduction, genetic instability.

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P.2.6: Valorisation of seafood canning industry residues for biosurfactants production

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Abstract:

The seafood canning industry plays a crucial role in the global supply of nutritious food with long shelf lives. Canning processes generate substantial amounts of solid and liquid residues. While the solids are used for feed and fish meal, valorisation of the liquid effluents is still limited. This study aimed to utilise the oily and the aqueous effluents from a cannery to produce mannosylerythritol lipids (MELs), glycolipids with high commercial potential owing to their surfactant and ceramide-like properties. In shake-flask experiments, the use of cannery oil effluent as a carbon source supported yeast growth and MELs production, with productivity of 2.19 ± 0.00 g_{MELs}/(L.day) and a purity of 0.9 $g_{MELs}/g_{(MELs+residual\ lipids)}$. The combined use of oil and aqueous effluents resulted in 2.06 ± 0.31 g_{MELs}/(L.day) with a purity of 0.7 $g_{MELs}/g_{(MELs+residual\ lipids)}$, decreasing process water intensity. Both conditions had higher productivities than the control using refined vegetable oil and fresh water (1.64 ± 0.16 g_{MELs}/(L.day), 0.8 purity). In a bioreactor operated in fed-batch mode, using a medium comprising both effluents, the productivity increased to 4.11 ± 0.12 g_{MELs}/(L.day). The MEL congener composition produced under these conditions was similar to that obtained in the control condition using refined vegetable oil and fresh water, with the main difference being the degree of fatty acid unsaturation, which appeared to be influenced by the unsaturation level of the substrate. This study highlights promising new pathways for valorisation of oily and aqueous side-streams from the canned seafood industry

Keywords: Mannosylerythritol lipids, biosurfactants, seafood cannery, circular economy, alternative substrates

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P 3.1 : Multi-cycle high-thoughput growth media optimization using batch bayesian optimisation

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Abstract:

Growth media composition is one of the highest-leverage decisions in any bioprocess, yet it remains dominated by Design of Experiments (DoE), a method that scales poorly with the number of components, struggles with the nonlinear nature of microbial growth, and routinely dismisses critical factors during early screening stages. As media complexity grows, the cost of suboptimal designs compounds: lost titer, wasted experiments, and slower time-to-process.

LABMaiTE's smart experiment design service replaces classical DoE with Batch Bayesian Optimization (BBO), a sequential, model-guided approach that uses Gaussian process surrogates and a local-penalization acquisition strategy to propose informative experiments in parallel. Rather than fixing factors after a single screening pass, BBO continues to weigh every component throughout the campaign, balancing exploration of the design space with exploitation of promising regions.

In a head-to-head wet-lab benchmark using Sporosarcina pasteurii on a fully automated microbioreactor platform (BioLector I, Opentrons OT-2, chi.bio turbidostat), BBO delivered a 28% higher biomass titer than a DoE-optimized medium at microscale. Critically, BBO identified iron as a limiting factor at high biomass, a component that DoE had dismissed as insignificant during screening. The advantage transferred to scale: in a 2 L stirred-tank bioreactor, the BBO medium reached an OD_{600} of 67.4, a 19% improvement over DoE and the highest batch density reported for S. pasteurii in the literature. These results position BBO as a practical, validated tool for accelerating bioprocess development.

Keywords: machine learning; Bayesian optimization; microbioreactor; media optimization, bioprocess development

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P 3.2: Local vs. Global: Achieving Better Raman Glucose Detection with Simplified Models

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Abstract:

Inline glucose monitoring is essential for reliable fermentation control. However, converting Raman spectra into accurate concentrations requires calibration models to link spectra to glucose levels. Conventional global models assume that a single calibration can handle all process variability, an assumption that often breaks down in industrial processes (Daniel Arturo Zavala-Ortiz et al. 2020; Daniel A. Zavala-Ortiz et al. 2020).

This work evaluates local models that adaptively select only the most relevant historical spectra for each prediction. Using six months of fermentation data, local modelling improved glucose prediction accuracy as evidenced by a RMSE decreased from 1.7 g/L to 1.4 g/L. Importantly, the local models remained stable throughout the entire six-month period despite natural batch to batch and temporal drift.

These results demonstrate that context specific calibration more effectively captures process variability than global approaches, strengthening the use of Raman based monitoring for advanced control, machine learning strategies, and future digital twins in bioprocessing.

Keywords: Raman ; chemometrics ; online analysis

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Zavala-Ortiz, Daniel A., Bruno Ebel, Meng-Yao Li, Dulce Ma. Barradas-Dermitz, Patricia M. Hayward-Jones, Maria G. Aguilar-Uscanga, Annie Marc, and Emmanuel Guedon. 2020. "Interest of Locally Weighted Regression to Overcome Nonlinear Effects during in Situ NIR Monitoring of CHO Cell Culture Parameters and Antibody Glycosylation." *Biotechnology Progress* 36(1):e2924. doi:10.1002/btpr.2924.

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P 3.3: Forecasting of Mannosylerythritol Lipids Production using Artificial Intelligence Methods

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Abstract:

Fermentations are complex and often unpredictable processes. However, fermentation-based bioprocesses generate large volumes of data that are currently underexplored. These data can be used to develop data-driven models, such as machine learning (ML) models, to improve process predictability. Among various fermentation products, biosurfactants have emerged as promising candidates for several industrial applications. Nevertheless, the large-scale production of biosurfactants is not yet cost-effective. This study aims to develop forecasting methods for the concentration of mannosylerythritol lipids (MELs), a type of biosurfactant, produced in Moesziomyces spp. cultivation. Three ML models, neural networks (NNs), support vector machines (SVMs), and random forests (RFs), were used. An NN provided predictions with a mean squared error (MSE) of 0.69 for day 4 and 1.63 for day 7 and a mean absolute error (MAE) of 0.58 g/L and 1.1 g/L, respectively. These results indicate that the model's predictions are sufficiently accurate for practical use, with the MAE showing only minor deviations from the actual concentrations. Both results are promising, as they demonstrate the possibility of obtaining reliable predictions of the MEL production on days 4 and 7 of fermentation. This, in turn, could help reduce process-related costs, enhancing its economic viability.

Keywords: Biosurfactant; Supervised learning; Fermentation; Prediction; Neural network;

References:

Vares, C. A., Agostinho, S. P., Fred, A. L. N., Faria, N. T., & Rodrigues, C. A. V. (2025). Machine Learning Strategies for Forecasting Mannosylerythritol Lipid Production Through Fermentation: A Proof-of-Concept. Applied Sciences, 15(7), 3709. https://doi.org/10.3390/app15073709

P 3.4: Silencing of functional genes circuit in *Bacillus subtilis* based on automated and reactive flow cytometry

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Abstract:

Many gene circuits in *Bacillus subtilis* offer useful functionalities for the development of biopesticides, notably sporulation and the secretion of bioactive compounds. The sporulation gene circuit is particularly critical to the quality of the final product. Other gene circuits could be engineered to trigger protein secretion upon contact with the target pest. However, these circuits must remain silenced during the main growth phase. It is well established that *Bacillus subtilis*, upon nutrient depletion, enters stationary phase and undergoes cellular differentiation, ultimately leading to the formation of dormant spores[1]. In continuous culture, low dilution rates drive cells into stationary phase[2], inducing sporulation, secretion of secondary metabolites, and release of extracellular proteins. Through genetic engineering, synthetic biology, and flow cytometry, it is possible to dynamically control activating or deactivating both sporulation and functionalized gene circuits[3].

In this work, we dynamically controlled the activation of the sporulation gene circuit in *Bacillus subtilis:PspoIIE_gfpmut2* using online flow cytometry measurements. When the percentage of sporulating cells in the bioreactor exceeded a defined threshold, glucose was automatically pulsed into the reactor to halt the sporulation process. The results demonstrate effective control over sporulation gene activation, resulting in a significant reduction in spore quantity compared to the uncontrolled system.

Keywords: *Bacillus subtilis*, sporulation, silencing, gene circuits, flow cell cytometry.

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P 3.5: Towards semi-automated image analysis for local bubble behavior

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Abstract:

Many bioprocesses depend on oxygen, and its low aqueous solubility makes bubble behavior crucial to understand. This behavior is affected by factors such as flow rate, power input, and viscosity. Numerous correlations for oxygen mass transfer and bubble size have been established over decades. However, these models typically rely on reactor-averaged measures and therefore fail to provide insights into local hydrodynamic gradients. To overcome this limitation, a pilot-scale reactor equipped with six probe inlets, positioned along the axial height, was used to acquire inline shadowgraphy images of gas bubbles. These images were analyzed using a CNN, enabling semi-automated, pixel-level characterization, as well as bubble shape construction. This provides an analysis of the individual bubble, including bubble size, shape, volume, and interfacial area, as well as an image-based determination of local gas holdup and mass transfer coefficient. The method was applied in an aqueous electrolyte solution, with increasing apparent viscosity, showing that bubble size increases overall as they rise through the reactor and that this increase depends strongly on the flow regime and viscous forces. This results in areas with a higher degree of gas bubbles, improving gas holdup. In water solutions, gas bubbles tend to be larger, and as viscosity increases, they break up more readily, thereby providing a greater interfacial area, which is essential for oxygen mass transfer. These results emphasize that bubble behavior is highly dynamic and must be understood to accurately predict and optimize oxygen mass transfer.

Keywords: Pilot scale, gas bubble dynamics, CNN, local mass transfer

P 5.1: A yeast-based nutrient enhancing growth of heme-auxotrophic bacteria for industrial fermentation

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Abstract:

Next-generation probiotics (NGPs) are emerging microorganisms identified through microbiota analyses that provide health benefits when administered appropriately. Many NGPs, such as *Faecalibacterium prausnitzii*, *Akkermansia muciniphila*, *Christensenella minuta*, and *Bacteroides spp*., play key roles in immune modulation, metabolic regulation, and gut barrier function, making them promising for applications in areas like metabolic disorders and oncology. They are strict anaerobes with complex nutritional requirements [1-2].

A specific challenge concerns heme-auxotrophic bacteria (HAB), which lack the full pathway for heme biosynthesis and therefore require external heme for growth [3-4]. Species such as *Bacteroides fragilis* and *Bacteroides uniformis* depend on heme supplementation due to incomplete porphyrin pathways [5]. To address this, Lallemand Bio-Ingredients developed an animal-free, yeast-based nutrient (Lalmedia™ PROTECT HB) designed for HAB and lactic acid bacteria.

In controlled experiments using 2L bioreactors and industrial media, Lalmedia™ PROTECT HB demonstrated superior performance compared to animal-derived heme for *B. uniformis* cultivation, confirming its effectiveness as an alternative.

Overall, Lalmedia™ PROTECT HB supports efficient industrial-scale production of NGPs, improving yields and downstream stability. It represents a sustainable and scalable solution for microbiome-based innovations, particularly for demanding anaerobic species.

Keywords: Heme, Yeast based nutrient, Next-generation probiotics, Lactic acid bacteria

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P 5.2: Exploring complex nitrogen consistency to ensure reproducible bioprocess manufacturing at scale

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Abstract:

Consistent raw material quality is essential for reproducible microbial fermentations. The impact of lot-to-lot variations is not always recognised or tested when scaling up or scaling down bioprocesses including during early development. Importance of raw material consistency becomes even more important when using complex sources, such as molasses for carbon or yeast derived bionutrients (YDBs) for nitrogen.

YDBs (such as yeast extract or yeast peptone) typically contain a plethora of compounds that fulfil the nutritional needs of a microbe of choice. A certain degree of variation is to be expected with any complex YDB but minimising such variations to ensure reproducible performance is vital for any fermentation process. Significant batch-to-batch variation in the composition of YDBs can lead to under- or over-supply of critical nutritional elements to cause yield losses, lower process productivity and even complete loss of production batches

In this study we will present chemo-physical and biological properties of more than 100 batches of YDBs, that were manufactured across multiple years, to demonstrate the expected levels of variance for different parameters such as total nitrogen, bioburden, colour etc. Focus will also be put on highlighting the impact of lot-to-lot variations on microbial performance via growth and functionality assays, using probiotic and starter culture strains. Overall the study will demonstrate a holistic view of the importance of incorporating batch-to-batch testing when scaling up or scaling down bioprocesses to ensure reproducibility in product output.

Keywords: yeast extract, yeast peptone, reproducibility, fermentation, bionutrients

P 5.3. Optimization and scale-up of fengycins production by fermentation in chemically defined or co-products based media

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Abstract:

Fengycins are cyclic lipopeptides naturally produced by *Bacillus subtilis* and recognized for their strong antifungal and surfactant properties. These properties lead to many interesting applications in the environmental, agricultural or health sectors. However, due to their current high production costs, fengycins are in most applications not able to challenge the chemically synthesized counterparts economically.

To improve their production yields, an integrated strain engineering strategy was implemented to increase basal fengycins production while suppressing the synthesis of competing lipopeptides. UV-induced mutagenesis combined with targeted genome editing using CRISPR-Cas9 enabled the selection of a high-producing *Bacillus subtilis* strain reaching production levels close to 1 g/L, with improved product specificity.

Following strain optimization, culture conditions were refined at bench scale to identify the most favorable production parameters. The optimized process was subsequently transferred to 200L bioreactors, enabling the production of several tens of grams of fengycins and demonstrating robust scalability up to the pilot scale. A downstream process combining successive filtration and precipitation steps was then developed, yielding fengycins preparations with purity levels exceeding 95%. The purified product was further characterized for its antifungal activity and physicochemical properties.

Overall, our results demonstrate the feasibility of large-scale fengycins production through an optimized and scalable bioprocess. We are now scaling up the fengycins production in a co-products based medium allowing to drastically reduce the production medium costs and make fengycins production more cost-efficient.

Keywords: Fengycins production, Strain engineering, Bioprocess scale-up, Upstream process, Downstream process

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