RESEARCH CENTRE
Paris

IN PARTNERSHIP WITH:
Institut Pasteur

2021
ACTIVITY REPORT

Project-Team

INBIO

Experimental and Computational Methods for Modeling Cellular Processes

IN COLLABORATION WITH: Centre de Bioinformatique, Biostatistique et Biologie Intégrative

DOMAIN

Digital Health, Biology and Earth

THEME

Modeling and Control for Life Sciences

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Project-Team INBIO

Creation of the Project-Team: 2019 November 01

Keywords

Computer sciences and digital sciences

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Other research topics and application domains

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1   Team members, visitors, external collaborators

Research Scientists

- Grégory Batt [Team leader, Inria, Senior Researcher, HDR]

- Jakob Ruess [Inria, Researcher]

Post-Doctoral Fellows

- Virgile Andreani [Institut Pasteur, until Jul 2021]

- Olivier Borkowski [Inria, until Sep 2021]

- Davin Lunz [Inria]

PhD Students

- Chetan Aditya [Inria and Institut Pasteur, until Oct 2021]

- Arthur Carcano [Université de Paris]

- Viktoriia Gross [Institut Pasteur]

- Sebastian Ramon Sosa Carrillo [Inria and Institut Pasteur]

Technical Staff

- François Bertaux [Institut Pasteur, Engineer, until Nov 2021]

- Achille Fraisse [Institut Pasteur, Engineer]

Visiting Scientist

- Lorenzo Pasotti [University of Pavia, part time visiting scientist]

External Collaborator

- François Bertaux [Lesaffre, from Dec 2021]

2   Overall objectives

The main objective of our research is to understand, control, and optimize cellular processes in single cells and at the population level. We combine experimental and theoretical work within a single team.

Our focus is on developing methods and models that take stochasticity of intracellular processes and heterogeneity of cell populations into account. To this end, we use both mixed-effects models as well as continuous-time Markov chains and their diffusion approximations. We develop methods for efficiently calculating with such models and use them to design optimally informative experiments and to reverse engineer unknown cellular processes from experimental data. Furthermore, we deploy models in order to optimally construct and optimally control synthetic gene circuits.

We have recently started to set up our own biology laboratory at Institut Pasteur. We develop novel experimental platforms that are designed to be fully automated, controllable by our own software, and capabale of updating the experimental plan in response to incoming measurements. Optogenetic actuation of intracellular processes, coupled to real time fluorescence measurements by microscopy or flow cytometry, then allows us to connect cellular processes with models and algorithms in real time.

The spritit of our work is that experimental platforms and circuits should be constructed with our theoretical work in mind, while our mathematical methods should be usable to adress concrete experimental questions in the lab.

3 Research program

3.1 Analysis and identification of stochastic (biochemical) reaction networks

The advancement of single-cell technologies in the last decades revealed that stochasticity is an inherent feature of cellular processes. Stochastic models, governed by the **chemical master equation** (CME), are widely used in applications to shed light on the functioning of biochemical reaction networks inside single cells. However, in most cases, the analysis of such models is based exclusively on Gillespie's stochastic simulation algorithm (SSA). SSA allows one to easily forward simulate the model but cannot be used very well for many important model analysis tasks. To overcome this problem, we develop various alternative approaches for calculating with stochastic models. In particular, we derive ordinary differential equations for the time evolutions of statistical moments of species abundances from the CME. We use **moment equations** and moment closure to develop methods for various model analysis tasks for which stochastic simulation is computationally too expensive to be practically useful, ranging from **parameter inference** to **model predictive control**. Furthermore, we recently started to make use of approximations of the CME with a **Fokker-Planck equation** to be able to also calculate with models for which low order statistical moments are not sufficient statistics of the full data and do not contain enough information.

3.2 Population dynamics emerging from randomness in single cells

Dynamics of cell populations growing in isolation or as part of some ecological system are often shaped by biochemical processes inside cells, for instance when these processes convey resistance to stressors or trigger cell fate decisions in reponse to environmental conditions. **Understanding how stochastic reaction events inside single cells affect emerging population dynamics**, and how selection effects at the population level feed back to shape single cell characteristics of cells in the population, is one of the key questions in biology. We develop **multi-scale modeling approaches** that allow us to derive emerging population dynamics from mechanistic descriptions of stochastic reaction networks inside single cells. In the past, we have used such approaches to study how stochasticity in restriction-modification systems, acting as simple bacterial innate immune systems, propagates to the ecology of bacteria and bacterial viruses and shapes the dynamics of bacterial populations. More generally, we develop and use these approaches in connection with experimental work in our lab for understanding and **controlling the dynamics of populations** in cases where **controllable system inputs inherently operate at the level of single cells** (e.g. optogenetics) but the output of interest is at the level of populations (e.g. bioproduction).

3.3 Optimal experimental design

One of the major problems in reverse engineering biochemical processes inside cells is that **cellular processes are high-dimensional and complex** with many unknown parameters while the **available data is low dimensional and corrupted by measurement errors**. Such problems can be alleviated by ensuring that the experimental plan is designed to yield data that provides as much information as possible about the unknown model parameters. We develop mathematical approaches and computational tools that can be used to calculate the expected amount of information that can be gained from a given experiment given a specification of either a stochastic model of the system (described above) or a deterministic model based on ordinary differential equations. These information calculation approaches are then coupled to optimization tools and used to plan **maximally informative experiments** in our applications.

3.4 Cybergenetics – real time control of biological processes

Cells have evolved uncountable numbers of feedback circuits to regulate their functionalities in the presence of changing environmental conditions. But can such feedback control also be externalized and placed under control of scientists? Early work on this topic suggested that **optogenetic systems, allowing for external regulation of gene expression**, have the potential to serve as an interface between cells and experimental platform that gives a computer the power to stir the functioning of cells via the application of light. We develop all the tools required to realize automated computer control of intracellular processes. On the experimental side, we develop yeast strains that are equipped with optogenetic promoters to

drive various functionalities. On the mathematical side, we develop models and software to equip our experimental platforms with the appropriate programs to realize sucessful feedback control, both at the level of single cells (microscopy) and at the level of populations (bioreactors and plate reader).

3.5 Platforms for automated reactive experiments

The core scientific activity of the team is to connect mathematical methods with biological applications in our lab. The interface between the two sides, that is the experimental platforms, is therefore of crucial importance for the success of our activities. However, platforms that can be purchased by vendors are typically delivered without the capacity to adapt the experimental plan in response to incoming measurements, a functionality that is crucially needed for deploying our computational methods (e.g. feedback control). Therefore, we develop novel experimental platforms and/or extend existing platforms with additional software and hardware that allows us to perform automated reactive experiments. Concretely, we develop a **microscopy platform and control software for yeast that uses a digital micromirror device to expose single cells to targeted light signals** that can be adjusted in real time in response to measurements taken from the cell. Furthermore, we develop a platform of 16 parallel **small scale automated bioreactors**, each equipped with controllable LEDs to allow for optogenetic gene expression and long-term reactive experiments in tightly controlled conditions. Automation of the platform is achieved via a low-cost open-source **pipetting robot that samples all reactors to a benchtop cytometer** in which single cell gene expression is measured in all sampled cells of all reactors. Finally, we develop **software to take full control of a commercial plate reader with liquid injection capabilities** (Tecan Spark). This platform allows us to use a Raspberry Pi to pilot 96 parallel reactive experiments where optical density is used as a readout of bacterial growth.

4 Application domains

4.1 Preamble

Since most of our research is at the interface of mathematics and biology, there often is no clear split between mathematical reaseach objectives and applications. For instance, feedback control of gene expression is simultaneously a mathematical and an applied problem.

4.2 Understanding resistance and tolerance to antibiotic treatments

The non-susceptibility of pathogenic bacteria to antibiotic treatments is a major health problem. Bacteria might escape treatments in two ways: being resistant or being tolerant. Whereas resistant bacteria can multiply in presence of antibiotics, tolerant bacteria can merely survive. Yet, tolerance is increasingly recognized as a major player in treatment failure. In particular, an increasing fraction of commensal and pathogenic *E coli* bacteria express extended-spectrum β-lactamases and/or carbapenemases. When individual bacteria die as a consequence of antibiotic treatments, these enzymes are released and hydrolyze the antibiotic molecules in the environment, conveying a transient protection to the remaining bacteria that lasts until the enzymes are degraded themselves. Understanding how this collective antibiotic tolerance (CAT) shapes population dynamics is difficult yet important for **optimally killing bacterial populations**: when a second antibiotic dose is applied directly after a first dose it will not be effective since the antibiotics will be degraded by the enzymes released from bacteria killed after the first dose; when the second dose is applied too late the surviving bacterial population will have regrown to a large size. Our plate reader platform allows us to apply complex temporal patterns of antibiotic treatments to bacteria over nearly two days. Parallezing such treatments in the 96 well plates allows us to generate rich data sets and to calibrate population dynamics models that can be used to optimize temporal treatment plans. One of the applied objectives of our team is to use these capacities to study a collection of fully-sequenced clinical isolates treated with a broad range of clinically important antibiotics and grown in various media. Ideally, this will lead to an approach that can be used to assay tolerance to antibiotics in hospitals instead of, or in addition to, standard antibiotic susceptibility tests, detecting resistance.

4.3 Optimization of protein production in yeast

Many proteins are of technological or therapeutical importance. The yeast *S. cerevisiae* is an interesting organism for protein bioproduction since it combines a relatively fast growth rate with good capacities to perform post-translational modifications needed for protein maturation and full functionality. However, imposing a strong demand on protein production to the cell places a significant burden on its physiology, either at the protein production level or at the maturation and secretion levels. Using systems and synthetic biology approaches, we aim at **better understanding the origins of the production bottlenecks** and then using modeling and control approaches, we aim at finding **optimal control solutions** for bioproduction. Three different strategies are envisioned. In the first approach, bioproduction stress sensors are used to observe in real time the physiological state of the cell, and the demand is externally tuned based on the stress level of the cell population. In the second approach, the stress sensor is used to tune the response capacities of the cell to the external demand, thus creating an internal feedback loop. In the third approach, we control the fraction of the producing cells by engineering an artificial differentiation system that implements the partial differentiation of grower cells into producer cells. The optimization problem is then to find the optimum ratio based on the external environment of the cells.

5 Social and environmental responsibility

5.1 Footprint of research activities

A significant part of our daily research activities involves molecular biology work and consumes plasticware and various chemicals. We also work on lab automation and develop experimental platform to paralellize experiments. However, we work with small biorectors (15 to 50mL), so volumes of cell cultures remain very modest.

We also occasionally use a computer cluster, notably for optimization, but the jobs remain relatively modest on a yearly basis.

5.2 Impact of research results

Regarding biological developments, we have two main research directions.

The first one deals with the optimization of bioproduction. Bioproduction is a domain of strategic importance. The field is higly technological and rapidly growing at the global scale. The market for biopharmaceuticals alone, that notably include vaccines and monoclonal antibodies, is estimated to $400B to $500B. France imports > 70% of its vaccines and > 95% of its monoclonal antibodies and lacks sovereignty. Therefore this field has a strong social, medical and economical importance.

The second research direction deals with antibiotic stewardship. The spread of antimicrobial resistance is a both a health and an ecological problem of global impact. Antibiotic stewardship aims at using these drugs in more appropriate ways. To do so, one has to better understand and quantify bacterial response to antibiotic treatments.

Therefore our two main research directions are both tightly connected with important health and social issues.

6 New software and platforms

We have continued our developments on MicroMator for enabling microscopy experiments to be reactive, on ReacSight for the automation of bioreactor-based platforms, and on PlateRider for the programmatic use of plate readers. For MicroMator and ReacSight, publications have been finalized, submitted and revised, and we are confident that they will be published in 2022. On the PlateRider side, publications are less advanced.

7 New results

7.1 A light tunable differentiation system for the creation and control of consortia in yeast

Participants: Chetan Aditya, François Bertaux, Gregory Batt, Jakob Ruess.

Artificial microbial consortia seek to leverage division-of-labour to optimize function and possess immense potential for bioproduction. Co-culturing approaches, the preferred mode of generating a consortium, remain limited in their ability to give rise to stable consortia having finely tuned compositions. In [1], we present an artificial differentiation system in budding yeast capable of generating stable microbial consortia with custom functionalities from a single strain at user-defined composition in space and in time based on optogenetically-driven genetic rewiring. Owing to fast, reproducible, and light-tunable dynamics, our system enables dynamic control of consortia composition in continuous cultures for extended periods. We further demonstrate that our system can be extended in a straightforward manner to give rise to consortia with multiple subpopulations. Our artificial differentiation strategy establishes a novel paradigm for the creation of complex microbial consortia that are simple to implement, precisely controllable, and versatile to use.

7.2 Using single-cell models to predict the functionality of synthetic circuits at the population scale

Participants: Chetan Aditya, François Bertaux, Gregory Batt, Jakob Ruess.

Mathematical modeling has become a major tool to guide the characterization and synthetic construction of cellular processes. However, models typically lose their capacity to explain or predict experimental outcomes as soon as any, even minor, modification of the studied system or its operating conditions is implemented. This limits our capacity to fully comprehend the functioning of natural biological processes and is a major roadblock for the de novo design of complex synthetic circuits. In [11], using a specifically constructed yeast optogenetic differentiation system as an example, we show that a simple deterministic model can explain system dynamics in given conditions but loses validity when modifications to the system are made. On the other hand, deploying theory from stochastic chemical kinetics and developing models of the system's components that simultaneously track single-cell and population processes allows us to quantitatively predict emerging dynamics of the system without any adjustment of model parameters. We conclude that carefully characterizing the dynamics of cell-to-cell variability using appropriate modeling theory may allow one to unravel the complex interplay of stochastic single-cell and population processes and to predict the functionality of composed synthetic circuits in growing populations before the circuit is constructed.

7.3 Beyond the chemical master equation: stochastic chemical kinetics coupled with auxiliary processes

Participants: Davin Lunz, Gregory Batt, Jakob Ruess.

The chemical master equation and its continuum approximations are indispensable tools in the modeling of chemical reaction networks. These are routinely used to capture complex nonlinear phenomena such as multimodality as well as transient events such as first-passage times, that accurately characterise a plethora of biological and chemical processes. However, some mechanisms, such as heterogeneous cellular growth or phenotypic selection at the population level, cannot be represented by

the master equation and thus have been tackled separately. In [2], we propose a unifying framework that augments the chemical master equation to capture such auxiliary dynamics, and we develop and analyse a numerical solver that accurately simulates the system dynamics. We showcase these contributions by casting a diverse array of examples from the literature within this framework and applying the solver to both match and extend previous studies. Analytical calculations performed for each example validate our numerical results and benchmark the solver implementation.

7.4 External control of microbial populations for bioproduction: A modeling and optimization viewpoint

Participants: François Bertaux, Jakob Ruess, Gregory Batt.

When engineering microbes for bioproduction, one is necessarily confronted with the existing tradeoff between efficient bioproduction and maintenance of the cell physiology and growth. Moreover, because cellular processes at the single-cell level are coupled with population dynamics via selection mechanisms, this question should be investigated at the population level. Identifying the temporal induction profile that maximizes production in the long term is highly challenging. External control allows to dynamically adapt the strength of the induction from the outside based on intracellular readouts. It allows benchmarking various regulation functions and, coupled with modeling approaches, identifying and applying optimal strategies. In [4], we review recent advances using quantitative approaches, modeling, and control theory that pave the way to compute external stimulations maximizing long-term production.

7.5 MicroMator: Open and Flexible Software for Reactive Microscopy

Participants: Zach Fox, Steven Fletcher, Achille Fraisse, Chetan Aditya, Sebastian Sosa-Carrillo, François Bertaux, Jakob Ruess, Gregory Batt.

Microscopy image analysis has recently made enormous progress both in terms of accuracy and speed thanks to machine learning methods. This greatly facilitates the online adaptation of microscopy experimental plans using real-time information of the observed systems and their environments. In [14], we report MicroMator, an open and flexible software for defining and driving reactive microscopy experiments, and present applications to single-cell control and single-cell differentiation.

7.6 Parameter inference for stochastic biochemical models from perturbation experiments parallelised at the single cell level

Participants: Andela Davidović, Gregory Batt, Jakob Ruess.

Understanding and characterising biochemical processes inside single cells requires experimental platforms that allow one to perturb and observe the dynamics of such processes as well as computational methods to build and parameterise models from the collected data. Recent progress with experimental platforms and optogenetics has made it possible to expose each cell in an experiment to an individualised input and automatically record cellular responses over days with fine time resolution. However, methods to infer parameters of stochastic kinetic models from single-cell longitudinal data have generally been developed under the assumption that experimental data is sparse and that responses of cells to at most a few different input perturbations can be observed. In [13], we investigate and compare different approaches for calculating parameter likelihoods of single-cell longitudinal data based on approximations of the chemical master equation (CME) with a particular focus on coupling the linear noise approximation (LNA) or moment closure methods to a Kalman filter. We show that, as long as cells are measured sufficiently frequently, coupling the LNA to a Kalman filter allows one to accurately approximate likelihoods

and to infer model parameters from data even in cases where the LNA provides poor approximations of the CME. Furthermore, the computational cost of filtering-based iterative likelihood evaluation scales advantageously in the number of measurement times and different input perturbations and is thus ideally suited for data obtained from modern experimental platforms. To demonstrate the practical usefulness of these results, we perform an experiment in which single cells, equipped with an optogenetic gene expression system, are exposed to various different light-input sequences and measured at several hundred time points and use parameter inference based on iterative likelihood evaluation to parameterise a stochastic model of the system.

7.7   A model-based approach to characterize enzyme-mediated response to antibiotic treatments: going beyond the SIR classification

Participants: Virgile Andréani, Gregory Batt.

To design appropriate treatments, one must be able to characterize accurately the response of bacteria to antibiotics. When exposed to β-lactam treatments, bacteria can be resistant and/or tolerant, and populations can exhibit resilience. Disentangling these phenomena is challenging and no consolidated understanding has been proposed so far. Because these responses involve processes happening at several levels, including the molecular level (e.g. antibiotic degradation), the cell physiology level (filamentation) and the population level (release of β-lactamases into the environment), quantitative modelling approaches are needed. In [12], we propose a model of bacterial response to β-lactam treatments that accounts for bacterial resistance, tolerance, and population resilience. Our model can be calibrated solely based on optical density readouts, can predict the inoculum effect, and leads to a mechanistically relevant classification of bacterial response to treatments that goes beyond the classical susceptible / intermediate / resistant classification. Filamentation-mediated tolerance and collective enzyme-mediated antibiotic degradation are essential model features to explain the complex observed response of cell populations to antibiotic treatments.

8   Partnerships and cooperations

8.1   International initiatives

- **ANR-FWF CyberCircuits** (2018-2022), on "Cybergenetic circuits to test composability of gene networks", co-coordinated by C. Guet (IST Austria, Austria) and Jakob Ruess.

  The objective of the Cybercircuit project is to explain and better predict how composed circuits function in vivo. To tackle this long standing question, we will construct hybrid bio-digital circuits in which a part of a network is effectively implemented as a biological genetic network whereas another part exists only virtually in the formof a model in a computer.

8.2   International research visitors

Lorenzo Pasotti, assistant professor at the Department of Electrical, Computer and Biomedical Engineering and at the Centre for Health Technologies of the University of Pavia (Italy) has been invited for three months in the InBio team.

8.3   European initiatives

- **FET Open COSY-BIO** (2017-2021), on "Control Engineering of Biological Systems for Reliable Synthetic Biology Applications", coordinated by Diego di Bernardo (Tigem), with Filippo Menolascina (Edinburgh U), Mario di Bernardo (Naples U) , Pascal Hersen (Institut Curie), Mustafa Khammash (ETHZ), Grégory Batt, Guy-Bart Stan (Imperial College), and Lucia Marucci (Bristol U).

The main objective of COSY-BIO is to identify generally applicable approaches to design closedloop feedback controllers for biological systems. The project will rely on control engineering for physical systems and on the exploitation of the unique features of living organisms.

8.4 National initiatives

8.4.1 ANR Projects

- **ANR MEMIP** (2016-2021) on "Mixed-Effects Models of Intracellular Processes", coordinated by G. Batt, with P. Hersen (Institut Curie), E. Cinquemani (IBIS, Inria) and M. Lavielle (XPOP, Inria/CNRS/Polytechnique).

  The objective of the MEMIP project is to develop mathematical methods for the identification of single-cell models, and to implement and bendchmark them on experimental data generated by innovative experimental platforms.

- **ANR CoGEx** (2016-2021) on "Computer Aided Control of Gene Expression", coordinated by P. Hersen (Institut Curie), with G. Batt and G. Truan (LISBP, CNRS/INSA).

  The CoGEx project aims at developing experimental and theoretical tools for the computer-based remote-control of live cells, and to use such a system to interrogate cellular processes at the single cell level.

- **Institut de Convergence Inception** (2016-2025) on the "Emergence of Diseases in Populations and in Individuals", coordinated by T. Bourgeron (Institut Pasteur). Partner institutes include Institut Pasteur, Paris Sciences et Lettres, Université de Paris, AP-HP, and research teams from CEA, CNRS, INSERM and INRA.

  The Inception's goal is to develop a core structure to mobilize data resources, numerical sciences, and fundamental experimental biology in a range of health issues. It uses integrative biology, social science and data science to understand the emergence of diseases in populations and in individuals.

- **PPR Antibiorésistance Anoruti** (2021-2025) on the "Analysis of non-response to antibiotics in vivo: application to Escherichia coli urinary tract infections", coordinated by I. El Meouche (Inserm).

  The objective of Anoruti is to identify the different factors involved in the fact that some bacteria sensitive to an antibiotic in vitro do not respond to treatment in vivo.

- **PPR Antibiorésistance Seq2Diag** (2021-2026) on "Whole genome sequencing and artificial intelligence to characterize and diagnose antibiotic resistance and capacity to escape treatment", coordinated by P. Glaser (Institut Pasteur).

  Genomic sequencing has revolutionized microbiological surveillance and molecular epidemiology. The objective of the Seq2Diag project is to provide a proof of concept for its use in hospital and veterinary laboratories as a diagnostic tool for in silico antibiotic sensitivity testing.

8.4.2 Inria Project Labs

- **IPL COSY** (2017-2021) on "Real-time control of synthetic microbial communities", coordinated by Eugenio Cinquemani (Ibis, Inria), with Jean-Luc Gouzé (Biocore, Inria), Grégory Batt, Frédéric Bonnans (Commands, Inria), Efimov Denis (Non-A, Inria), and Hans Geiselmann (BIOP, Université Grenoble-Alpes), Béatrice Laroche (Maiage, Inra Jouy-en-Josas).

  The COSY project aims at developping automated experimental platforms and control methods for heterogeneous microbial populations to propose solutions for the optimization of bioproduction processes.

9 Dissemination

9.1 Promoting scientific activities

9.1.1 Scientific events: organisation

In 2021, the working group Biologie Systémique Symbolique (BIOSS) organized a series of monthly seminars in lieu of its usual yearly working days. In 2021, Gregory Batt, a member of the steering committee, helped with their organizations.

9.1.2 Scientific events: selection

Jakob Ruess was a reviewer for the European Control Conference (ECC 2021).

9.1.3 Journal

Gregory Batt was a reviewer for reviewer for ACS Synthetic Biology.
   Jakob Ruess was a reviewer for Bioinformatics and The FEBS Journal.

9.1.4 Invited talks

Gregory Batt gave an invited talk entitled "Methods and tools for the quantitative characterization of engineered biomolecular systems", at the Pasteur Qbio symposium Bridging the scales in June 2021, online. He also gave an invited seminar at the Physical Microfluidics and Bioengineering group in Feb 2021, online.

9.1.5 Leadership within the scientific community

Gregory Batt is a member of the scientific board of the French research network on Bioinformatics (GdR BIM) and a member of the steering committee of the working group on Symbolic Systems Biology (GT Bioss), affiliated to two French research networks, on Bioinformatics (GdR BIM) and on Mathematical computer science (GdR IM).

9.1.6 Scientific expertise

Gregory Batt has been a reviewer for the PhD thesis of Federica Cella of Istituto Italiano di Tecnologia/Università di Genova.
   Jakob Ruess has been a member of the thesis advisory committee of Emrys Reginato at Université Grenoble-Alpes/INRIA, Grenoble.
   Gregory Batt has been a member of the Comité Scientifique Stratégique du Grand Défi "Biomédicaments: améliorer les rendements et maîtriser les coûts de production", and of the selection committees for the ANR call "Nouveaux Systèmes d'Expression" and for the BPI call on Bioproduction.
   Gregory Batt has also been a member of the selection committee for the Pasteur Transverse Research Programs. He has also been involved in the selection committee for junior group leaders at Pasteur.

9.1.7 Research administration

Gregory Batt is the deputy director of the department of Computational Biology at Institut Pasteur.

9.2 Teaching - Supervision - Juries

9.2.1 Teaching

- Grégory Batt (30h), Jakob Ruess (15h), Computational Biology, M1, Master Interdisciplinary Approaches to Life Sciences (AIRE-LiSc).

- François Bertaux (12h), Systems Biology, M1, Master Interdisciplinary Approaches to Life Sciences (AIRE-LiSc).

- Olivier Borkowski (5h), Synthetic Biology, M2 Master AgroParisTech.

9.2.2 Supervision

Three PhD students defended their thesis this year

- PhD defended: Chetan Aditya, "Control of heterogenous synthetic microbial systems", ED FIRE, Université de Paris, Started in Feb. 2018 and defended in Oct 2021. Supervision by Grégory Batt with Jakob Ruess.

- PhD defended: Arthur Carcano, "Iterative design of single-cell experiments to learn single-cell models of biological systems", ED FIRE, Université de Paris. Started in Oct. 2018 and defended in Dec 2021. Supervision by Jakob Ruess and Grégory Batt.

- PhD defended: Sebastian Sosa Carrillo, "Understanding the cost of protein production in yeast", ED FIRE, Université de Paris. Started in Feb. 2018 and defended in Dec 2021. Supervision by Grégory Batt with François Bertaux.

- PhD in progress: Viktoriia Gross, "An integrative approach to characterize the two sides of enzyme mediated antibiotic escape: resistance and tolerance. Started in Oct. 2020. Supervision by Imane ElMeouche, Erick Denamur and Grégory Batt.

10   Scientific production

10.1   Major publications

[1] C. Aditya, F. Bertaux, G. Batt and J. Ruess. 'A light tunable differentiation system for the creation and control of consortia in yeast'. In: *Nature Communications* 12 (5th Oct. 2021), p. 5829. DOI: 10.1038/s41467-021-26129-7. URL: https://hal-pasteur.archives-ouvertes.fr/pasteur-03263577.

[2] D. Lunz, G. Batt, J. Ruess and J. Frédéric Bonnans. 'Beyond the chemical master equation: stochastic chemical kinetics coupled with auxiliary processes'. In: *PLoS Computational Biology* (28th July 2021). DOI: 10.1371/journal.pcbi.1009214. URL: https://hal.inria.fr/hal-02991103.

10.2   Publications of the year

International journals

[3] C. Aditya, F. Bertaux, G. Batt and J. Ruess. 'A light tunable differentiation system for the creation and control of consortia in yeast'. In: *Nature Communications* 12 (5th Oct. 2021), p. 5829. DOI: 10.1038/s41467-021-26129-7. URL: https://hal-pasteur.archives-ouvertes.fr/pasteur-03263577.

[4] F. Bertaux, J. Ruess and G. Batt. 'External control of microbial populations for bioproduction: A modeling and optimization viewpoint'. In: *Current Opinion in Systems Biology* 28 (Dec. 2021), p. 100394. DOI: 10.1016/j.coisb.2021.100394. URL: https://hal.inria.fr/hal-03536479.

[5] D. Lunz. 'On rapid oscillations driving biological processes at disparate timescales'. In: *Physical Biology* 18.3 (15th Mar. 2021), p. 036002. DOI: 10.1088/1478-3975/abd9db. URL: https://hal.inria.fr/hal-03287815.

[6] D. Lunz. 'Optimizing Noisy Complex Systems Liable to Failure'. In: *SIAM Journal on Applied Mathematics* 82.1 (5th Jan. 2022), pp. 25–48. DOI: 10.1137/21M1416126. URL: https://hal.inria.fr/hal-03537040.

[7] D. Lunz, G. Batt, J. Ruess and J. Frédéric Bonnans. 'Beyond the chemical master equation: stochastic chemical kinetics coupled with auxiliary processes'. In: *PLoS Computational Biology* (28th July 2021). DOI: 10.1371/journal.pcbi.1009214. URL: https://hal.inria.fr/hal-02991103.

Doctoral dissertations and habilitation theses

[8]   C. Aditya. 'Development of a light tunable differentiation system for the creation and control of microbial consortia in S. cerevisiae, its single cell characterization for development of predictive models, and application to heterologous expression'. Université de Paris, 1st Oct. 2021. URL: https ://hal.inria.fr/tel-03612653.

[9]   A. Carcano. 'Methods and tools for the optimal design of experiments applied to the efficient characterization of enzyme-mediated escape to antimicrobial treatments'. Université de Paris, 13th Dec. 2021. URL: https://hal.inria.fr/tel-03612649.

[10] S. Sosa-Carrillo. 'Pipeline for the systematic characterization of heterologous protein secretory load to assess bioproduction efficiency'. Université de Paris, 15th Dec. 2021. URL: https://hal.i nria.fr/tel-03612661.

Reports & preprints

[11] C. Aditya, F. Bertaux, G. Batt and J. Ruess. *Using single-cell models to predict the functionality of synthetic circuits at the population scale.* 4th Aug. 2021. DOI: 10.1101/2021.08.03.454887. URL: https://hal.inria.fr/hal-03544044.

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