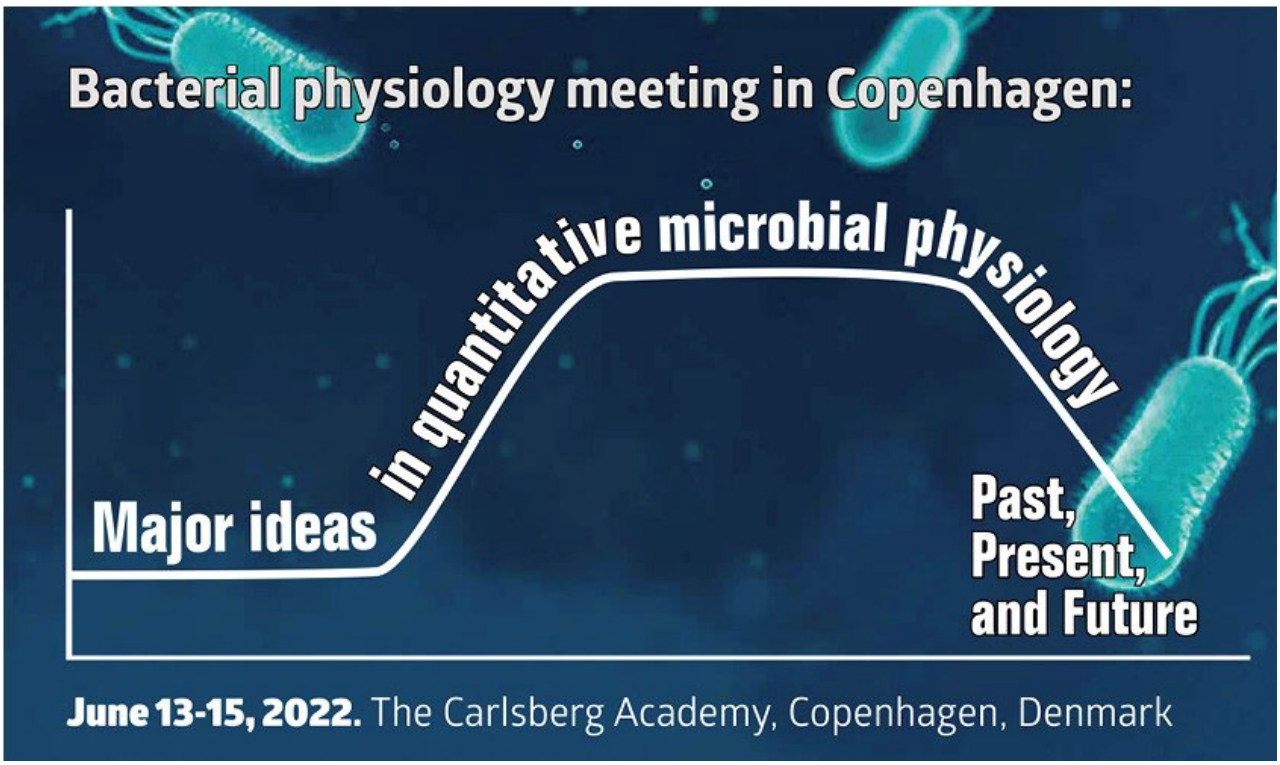
Program Book



The meeting is generously sponsored by the Novo Nordisk Foundation.

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Meeting venue:

Carlsberg Academy
Gamle Carlsberg Vej 15
DK-2500 Copenhagen

Wifi access:

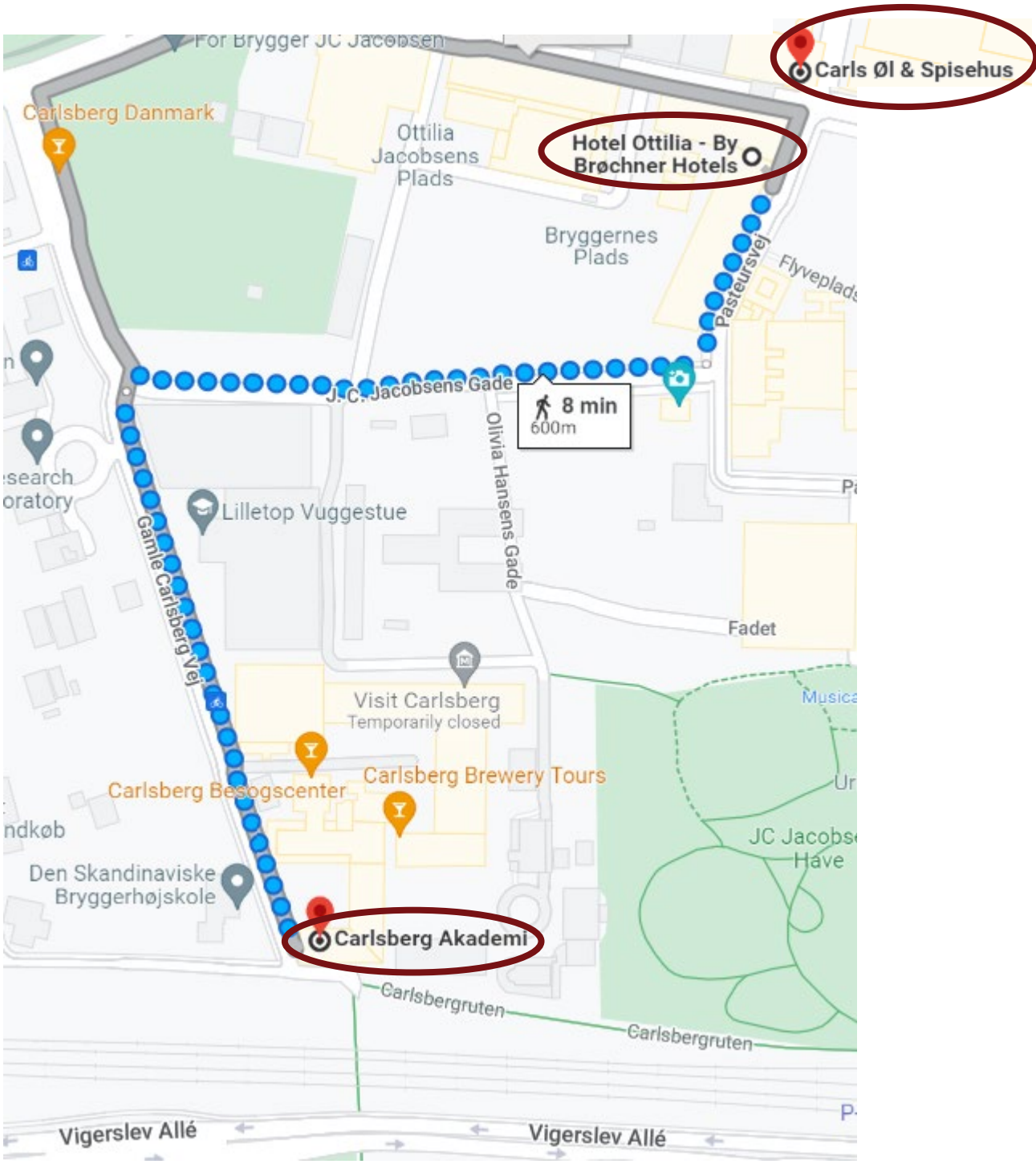
Network: Carlsberg Academy Guest
Password: Probably.

Conference Dinner, June 13

Carl's Øl & Spisehus
Bag Elefanterne 20
DK-1799 Copenhagen

Invited speakers/guests hotel:

Hotel Ottilia
Bryggernes Plads 7
DK-1778 Copenhagen



Day 1: Monday, June 13, 2022

8:15-9:00	Registration
9:00-9:20	**Welcome & opening remarks** Suckjoon Jun
9:20-10:20 **session chair:** **Suckjoon Jun**	**Terry Hwa** (20'+5') "Sculpting of the gene expression landscape by physiological constraints" **Stefan Klumpp** (12'+3') "Resource allocation in bacterial motility" **Severin Schink** (12'+3') "Quantitative insights into bacterial starvation survival "
10:20-11:05	**Tea/Coffee**
11:05-12:05 **session chair:** **Ala Trusina**	**Martin Lercher** (12'+3') "Parsimonious allocation of *E. coli* dry mass governs the enzyme-substrate relationship" **Nathalie Balaban** (20'+5') "The Impact of the Disrupted Cellular State on Antibiotic Response" **Rami Pugatch** (12'+3') "A unifying autocatalytic network-based framework for bacterial growth laws"
12:05-15:00	**Lunch + Poster Session 1 (even numbers)/ Free-form pioneers session 1**
15:00 - 16:20 **session chair:** **Anna Ebbensgaard**	**Petra Levin** (20'+5') “The environment shapes the cell” **Vahid Shahrezaei** (12'+3') "Mechanisms of size control across growth conditions in *E. coli* and Pombe" **Shila Banerjee** (20'+5'; remote) "Cell morphology as regulator of bacterial fitness and adaptation"
16:20-17:00	**Tea/Coffee**
17:00-18:10 **session chair:** **Mogens Kilstrup**	**Tanneke den Blaauwen** (20'+5') "Quantitative analysis of the timing of midcell localization of division proteins reveals specific roles for endopeptidases PBP4 and PBP7" **Marco C Lagomarsino** (12'+3') "Out-of-equilibrium behavior of microbial growth/division reveals basic regulatory mechanisms" **Willie Donachie** (20'+5'; remote) "Answering the Nordström Question (and maybe a few others)."
19:00	Conference dinner for all participants. Restaurant Carl's Øl & Spisehus

Day 2: Tuesday, June 14, 2022

8:30-9:00	Arrival
9:00-10:25 **session chair: Jakob Frimodt-Møller**	**Matthias Heinemann** (20'+5') "From physiology to moving molecules?" **Oleg Igoshin** (12'+3') "Making cell-fate-decisions: lessons from *B. subtilis*" **Alicia Berkvens** (12'+3') "Growth consequences of the inhomogenous organization of the bacterial cytoplasm" **Seungeun Oh** (20'+5') "Quantitative single-cell physiology of growth and size regulation using label-free microscopy"
10:25-11:00	**Tea/Coffee**
11:00-12:25 **session chair: Kim Sneppen**	**Yuichi Wakamoto** (20'+5') "Raman-omics correspondence reveals homeostatic core and stoichiometrically-balanced dynamical groups inside cells" **Michael Sørensen** (12'+3') "The degradation rate of ribosomes during stationary phase depends on the type of missing nutrient" **Yusuke Himeoka** (12'+3') "Emergence of growth and dormancy from a kinetic model of the *Escherichia coli* central carbon metabolism" **Johan Elf** (20'+5') Coordination of replication initiation to growth in *Escherichia coli*
12:25-15:00	**Group photo;** **Lunch and Poster Session 2 (odd numbers)/** **Freeform pioneers session 2**
15:00 - 16:10 **session chair: Namiko Mitarai**	**Søren Molin** (20'+5') “Investigations of microbial physiology in adapting bacterial populations involved in airway infections” **Sven van Teffelen** (12'+3') "Regulation of mass density in rod-shaped bacteria" **Gene-Wei Li** (20'+5') "Predictability and Unpredictability of Expression-Fitness Relationships"
16:10-16:40	**Tea/Coffee**
16:40-18:00 **session chair: Sine Lo Svenningsen**	**Susan Gottesman** (20'+5') "Coming and Going: *E. coli* circuitry for adjusting to and recovering from stress" **Thomas Julou** (12'+3') Growth-rate controls the sensitivity of gene regulatory circuits **Morten Nørholm** (12'+3') "Molecular mechanisms in the evolution of nutrient-limited bacterial colonies" **Megan Bergkessel** (12'+3') "Regulation of protein synthesis during growth arrest"
18:00-20:00	**Late-night poster session:** Reception-style buffet, the bar is open!

Day 3: Wednesday, June 15, 2022

8:30-9:00	Arrival
9:00-10:30 **session chair:** **Flemming Hansen**	**Tsutomu Katayama** (20'+5'; remote) Dynamic mechanisms constituting regulation for the replication initiator DnaA protein in *Escherichia coli* **Godefroid Charbon** (12'+3') Arresting chromosome replication upon energy starvation in *Escherichia coli* **Mareike Berger** (12'+3') Robust replication initiation from coupled homeostatic mechanisms **Steen Pedersen** (20'+5') The genetic code, in addition to encode the amino acids, also encodes signals that modify gene expression by affecting mRNA half-life and premature transcription termination.
10:30-11:00	**Tea/Coffee**
11:00-12:00 **session chair:** **Tove Atlung**	**Teuta Pilizota** (20+5') "The proton motive force determines *Escherichia coli*'s robustness to extracellular pH" **Claudia Igler** (12'+3') "Control of phage lysis timing through host cell physiology" **Hanne Ingmer** (12'+3') "Transduction in shaping bacterial genomes"
12:00-13:45	**Lunch**
13:45 - 15:10 **session chair:** **Frank Bruggeman**	**Jens Nielsen** (20'+5') Systems biology of yeast metabolism **Karin Mitosch** (12'+3') "Isotope tracing reveals metabolic activities of Salmonella during intracellular infection" **Einat Segev** (12'+3', remote) "Lag Phase Shortening in Bacteria" **Roberto Kolter** (20'+5', remote) "Thoughts on the Physiology of Natural Bacterial Populations."
15:10-15:40	**Tea/Coffee**
15:40-16:50 **session chair:** **Arieh Zaritsky**	**Akos Kóvacs** (20'+5') "Multi-species biofilm and the interaction within" **Kathryn Turnbull** (12'+3') Structural and function studies of bacterial ABCF ribosome associated resistance factors" **Stephen Cooper** (20'+5'; remote) "Three Critical Manifestos for the Cell Cycle: On Synchronization, on G0, on Gene Expression"
16:50-17:00	Closing

List of participants

Name	Email Address	Institution	Participation
Ákos Kovács	atkovacs@dtu.dk	Technical University of Denmark	invited speaker
Ala Trusina	trusina@nbi.ku.dk	University of Copenhagen	poster presentation
Alba García Vázquez	alba.vazquez@nbi.ku.dk	University of Copenhagen	poster presentation
Alfonso Soler Bistue	asoler@iib.unsam.edu.ar	Universidad Nacional de San Martín / Consejo Nacional de Investigaciones Científicas y Técnicas (UNSAM/CONICET)	poster presentation
Alicia Berkvens	a.m.berkvens@vu.nl	Amsterdam Institute of Molecular and Life Sciences	oral presentation
Andreas Berglund Nielsen	andreas.berglund@nbi.ku.dk	University of Copenhagen	poster presentation
Anna Ebbensgaard	anna.ebbensgaard@bio.ku.dk	University of Copenhagen	session chair
Anna Knöppel	annaknoppel@gmail.com	ICM, Uppsala University	poster presentation
Ardré Maxime	maxime.ardre@espci.psl.eu	ESPCI Paris	poster presentation
Arieh Zaritsky	ariehzar@gmail.com	Ben-Gurion University of the Negev	special guest
Benjamin Roller	benjamin.robert.roller@univie.ac.at	University of Vienna	poster presentation
Bertil Gummesson	bertil.gummesson@bio.ku.dk	University of Copenhagen	assisting organizer
Charles Helmstetter	chelmste@fit.edu	Florida Institute of Technology	special guest, remote
Christoph Weigel	christoph.weigel.berlin@gmail.com	Technical University of Berlin	participant
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Elio Schaechter	elios179@gmail.com	Tufts University	special guest, remote
Ferdinand Greiss	ferdinand.greiss@weizmann.ac.il	Weizmann Institute of Science	poster presentation
Flemming G. Hansen	fgha@dtu.dk	Technical University of Denmark	special guest
Frank Bruggeman	f.j.bruggeman@vu.nl	Vrije University Amsterdam	session chair
Gene-Wei Li	gwli@mit.edu	Massachusetts Institute of Technology	invited speaker
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Karin Hammer	kha@bio.dtu.dk	Technical University of Denmark	special guest
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Kaspar von Meyenburg	k.v.meyenburg@schipf.ch	Schipf Weingut am Zürichsee	special guest
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Laura Martínez Alvarez	laura.martinez@bio.ku.dk	University of Copenhagen	participant
Lise Goltermann	lisgol@biosustain.dtu.dk	Technical University of Denmark	participant

List of participants, continued

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Mareike Berger	m.berger@amolf.nl	AMOLF, Amsterdam	oral presentation
Martin Lercher	martin.lercher@hhu.de	Heinrich Heine University Düsseldorf	oral presentation
Matthias Heinemann	m.heinemann@rug.nl	University of Groningen	invited speaker
Megan Bergkessel	mbergkessel001@dundee.ac.uk	University of Dundee	oral presentation
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Michael Manhart	michael.manhart@env.ethz.ch	ETH Zürich	poster presentation
Mikkel Svenningsen	mikkel.svenningsen@nbi.ku.dk	University of Copenhagen	poster presentation
Mireia Cordero	mireia.sanchez@nbi.ku.dk	University of Copenhagen	poster presentation
Mogens Kilstrup	mki@bio.dtu.dk	Technical University of Denmark (DTU)	poster presentation
Mohammad Roghanian	mohammad.roghanian@regionh.dk	Rigshospitalet, Copenhagen	poster presentation
Morten Nørholm	morno@biosustain.dtu.dk	Technical University of Denmark	oral presentation
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Namiko Mitarai	mitarai@nbi.dk	University of Copenhagen	organizer
Nathalie Balaban	nathalie.balaban@mail.huji.ac.il	Hebrew University of Jerusalem	invited speaker
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Ole Skovgaard	olesk@ruc.dk	Roskilde University	poster presentation
Oleg Igoshin	igoshin@rice.edu	Rice University	oral presentation
Petra Levin	plevin@wustl.edu	Washington University	invited speaker
Philip Hanawalt	hanawalt@stanford.edu	Stanford University	special guest, remote
Pranas Grigaitis	p.grigaitis@vu.nl	Vrije Universiteit Amsterdam	poster presentation
Ralf Steuer	steuer.ralf@gmail.com	Humboldt-University of Berlin	poster presentation
Rami Pugatch	rami.pugatch@gmail.com	Ben-Gurion University of the Negev	oral presentation
Roberto Kolter	roberto_kolter@hms.harvard.edu	Harvard Medical School	invited speaker, remote
Rocio Espinosa	rocio.espinosa@bio.ku.dk	University of Copenhagen	poster presentation
Seungeon Oh	seungeun_oh@hms.harvard.edu	Harvard Medical School	invited speaker
Severin Schink	severin_schink@hms.harvard.edu	Harvard Medical School	oral presentation
Sidsel Henriksen	sidsel.henriksen@bio.ku.dk	University of Copenhagen	assisting organizer
Sine Lo Svenningsen	sls@bio.ku.dk	University of Copenhagen	organizer
Shiladitya Banerjee	shiladtb@andrew.cmu.edu	Carnegie Mellon University	invited speaker, remote
Stanley Brown	stanley@nbi.ku.dk	University of Copenhagen	participant
Steen Pedersen	steenp@fastmail.com	University of Copenhagen	invited speaker
Stefan Klumpp	stefan.klumpp@phys.uni-goettingen.de	University of Göttingen	oral presentation
Stephen Cooper	cooper@umich.edu	University of Michigan Medical School	invited speaker, remote
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Susan Gottesman	gottesms@helix.nih.gov	National Cancer Institute, NIH	invited speaker
Susanne Häussler	susanne.haeussler@helmholtz-hzi.de	Rigshospitalet, Copenhagen	poster presentation
Sven van Teeffelen	sven.vanteeffelen@gmail.com	University of Montreal	oral presentation
Søren Molin	sm@bio.dtu.dk	Technical University of Denmark	invited speaker
Tanneke den Blaauwen	t.denblaauwen@uva.nl	University of Amsterdam	invited speaker
Terence Hwa	hwa@ucsd.edu	UC San Diego	invited speaker
Teuta Pilizota	teuta.pilizota@ed.ac.uk	University of Edinburgh	invited speaker
Thias Oberg Boesen	thias.boesen@bio.ku.dk	University of Copenhagen	participant
Thomas Julou	thomas.julou@unibas.ch	University of Basel	oral presentation
Thomas S. Stenum	thomas.stenum@icm.uu.se	Uppsala University	poster presentation
Tove Atlung	atlung@ruc.dk	Roskilde University	special guest
Tsutomu Katayama	katayama@phar.kyushu-u.ac.jp	Kyushu University	invited speaker, remote
Vahid Shahrezaei	v.shahrezaei@imperial.ac.uk	Imperial College London	oral presentation
William Donachie	uilleam4@gmail.com	University of Edinburgh	invited speaker, remote
Yong Everett Zhang	yong.zhang@bio.ku.dk	University of Copenhagen	poster presentation
Yuichi Wakamoto	cwaka@mail.ecc.u-tokyo.ac.jp	University of Tokyo	invited speaker
Yusuke Himeoka	yhimeoka@g.ecc.u-tokyo.ac.jp	University of Tokyo	oral presentation

List of speakers (alphabetical)

Ákos Kovács
Technical University of Denmark
Multi-species biofilm and the interaction within

Alicia Berkvens
Amsterdam Institute of Molecular and Life Sciences
Growth consequences of the inhomogenous organization of the bacterial cytoplasm

Claudia Igler
ETH Zurich
Control of phage lysis timing through host cell physiology
Abstract:
Bacteriophages are viruses that infect bacterial cells to use the host cell machinery for their own replication and assembly of progeny virions. The release of these virions, and therefore phage fitness, generally depends on lysis of the bacterial cell at a defined timing, which is determined by phage lysis proteins. How changes in environmental conditions affect phage lysis timing through the physiological state of the host remains little understood. Using single-cell microscopy and mathematical modelling, we investigated how the lysis timing distribution across a bacterial population changes with environmental conditions. For the lysis system of phage λ, we find that lysis time is longer at faster growth of the host. This can be explained through faster protein dilution via cell division. Lytic replication of phage λ however usually arrests cell division, leading to the opposite effect, with faster growth leading to faster lysis with very low variation. Hence, individual phage functions are likely to be dependent on the physiological state of the host, and it might be beneficial for phages to carry genes that moderate this dependence."

Einat Segev
Weizmann Institute of Science
Lag Phase Shortening in Bacteria
Abstract:
Most heterotrophic bacteria in the ocean rely on metabolites that are produced by photosynthetic hosts. In the nutrient-poor marine environment, heterotrophic bacteria experience prolonged phases of starvation. When these bacteria encounter a nutrient source, they have to rapidly respond and adjust their metabolism. An early response is vital to outcompete co-occurring bacteria.
Bacteria adjust their metabolism in response to newly encountered nutrients during the lag phase. During lag phase, cell division does not occur and bacteria synthesize enzymes needed to produce essential building blocks and energy. Faster adjustment of the cellular machinery to available nutrients during lag phase allows earlier transition to exponential growth.
Interestingly, various marine bacteria specialize in an early response to micro-algal exudates which are a key nutrient source in the marine environment. What allows these bacteria to rapidly respond to algal exudates?
To understand the bacterial physiology that underlies an early response to algal exudates, we have studied a model marine bacterium called Phaeobacter inhibens. P. inhibens and its relatives from the Roseobacter group naturally interact with various micro-algae and often dominate algal-associated niches.
By studying the physiology of a marine bacterium under culture conditions that are inspired by its ecology, we revealed that bacteria shorten their lag phase in response to algal exudates. Specifically, we found that bacteria harvest methyl groups from methylated algal compounds. These methyl groups are used both for energy production, and are also fuelled into the methionine cycle. The incorporation of the harvested methyl groups into the methionine cycle results in lag phase shortening. Genetic and biochemical characterization unveil a central bacterial enzyme responsible for lag phase shortening. Furthermore, we show that similar mechanisms for expediting the lag phase are found in various bacteria from diverse environments."

Gene-Wei Li
MIT
Predictability and Unpredictability of Expression-Fitness Relationships

List of speakers, continued (alphabetical)

Godefroid Charbon
University of Copenhagen
Arresting chromosome replication upon energy starvation in *Escherichia coli*

Hanne Ingmer
University of Copenhagen
Transduction in shaping bacterial genomes.
Abstract:
Bacteriophage mediated transfer of bacterial DNA was previously considered to be either generalized or specialized. Recently, however, new transduction types have been discovered and in some bacteria the efficacy suggests that transduction may be a central component in shaping bacterial genomes. My talk will focus on transduction in the human pathogen, Staphylococcus aureus and I will discuss the role of prophages in efficient transfer of bacterial DNA located upstream and downstream of the phage."

Jens Nielsen
Bioinnovation Institute/Chalmers University of Technology
Systems biology of yeast metabolism

Johan Elf
Uppsala universitet
Coordination of replication initiation to growth in *Escherichia coli*

Karin Mitosch
EMBL Heidelberg
Isotope tracing reveals metabolic activities of Salmonella during intracellular infection

Kathryn Turnbull
Rigshospitalet
Structural and function studies of bacterial ABCF ribosome associated resistance factors

Marco Cosentino Lagomarsino
IFOM Foundation / University of Milan
Out-of-equilibrium behavior of microbial growth/division reveals basic regulatory mechanisms
Abstract:
Proliferating cells couple nutrient and environment sensing to both growth and cell-cycle progression, and a current challenge is quantifying how this task is precisely carried out. Such mechanistic aspects on how nutrient sensing affects the regulation of cell growth and division are only revealed when the equilibrium “balanced growth” conditions where phenomenological “growth laws” apply are perturbed. Focusing on the model organism *E. coli*, this talk will use mathematical models rooted in data to address the hierarchy of events that single cells undergo to respond to simple environmental changes. First, I will present a mechanistic theory of resource allocation, based on nutrient sensing and transcriptional regulation by ppGpp, and show that it generally predicts damped oscillatory response to perturbations. Second, a common assumption is that the biochemical circuits that sense nutrient levels adjust the rate of cell growth, which in turn affects the division machinery, determining cell division rate. Instead, I will show that controlled nutrient-shift experiments combining microscopy and microfluidics show that the growth and the division machinery sense nutrient levels in parallel, but the division machinery adapts faster, likely through the use of a nutrient-mediated threshold-accumulation circuit."

List of speakers, continued (alphabetical)

Mareike Berger

AMOLF

Robust replication initiation from coupled homeostatic mechanisms

Abstract:

The bacterium *Escherichia coli* initiates replication once per cell cycle at a precise volume per origin and adds an on average constant volume between successive initiation events, independent of the initiation size. Yet, a molecular model that can explain these observations has been lacking. Experiments indicate that *E. coli* controls replication initiation via titration and activation of the initiator protein DnaA. Here, we study by mathematical modelling how these two mechanisms interact to generate robust replication-initiation cycles. We first show that a mechanism solely based on titration generates stable replication cycles at low growth rates, but inevitably causes premature re-initiation events at higher growth rates. In this regime, the DnaA activation switch becomes essential for stable replication initiation. Conversely, while the activation switch alone yields robust rhythms at high growth rates, titration strongly enhances the stability of the switch at low growth rates. Our analysis thus predicts that both mechanisms are required to generate robust replication cycles at all growth rates. In addition, it reveals how an origin-density sensor yields adder correlations.

Martin Lercher

Heinrich Heine University Düsseldorf

Parsimonious allocation of *E. coli* dry mass governs the enzyme-substrate relationship, explaining the growth rate-dependencies of the biosynthetic protein sector and the composition of the translation machinery

Abstract:

Reaction fluxes are jointly determined through the abundances of catalysts and of their reactants. However, little is known about the relationships between those abundances. We hypothesized that enzyme-substrate relationships arise from the maximally parsimonious allocation of cellular dry mass. For a cellular reaction network composed of effectively irreversible reactions, maximal reaction flux is achieved when the dry mass allocated to each substrate is equal to the dry mass of the unsaturated (or “free”) enzymes waiting to consume it. This prediction is confirmed by available data for enzyme and substrate concentrations. The most expensive cellular catalysts in minimal media are the ribosome and MetE, which catalyzes the last step in Methionine synthesis. For these two, optimal dry mass allocation leads to near perfect predictions of their growth rate-dependent abundances. The corresponding organizing principle may also explain the apparent offset resulting from extrapolating the catabolic proteome sector abundance to zero growth rate, an important ingredient in models based on Bacterial Growth Laws. Moreover, it provides a quantitative explanation for the growth rate-dependent composition of the translation machinery, comprising not only the ribosome, but also charged tRNAs, mRNA, and elongation factors. Surprisingly, part of this growth rate-dependence is regulated through the relative genomic positioning of tRNAs and ribosomes. In sum, the organizing principle of parsimonious dry mass allocation appears to provide a fundamental rationale for the condition-dependent cellular investment into different types of molecules.

Matthias Heinemann

University of Groningen

From physiology to moving molecules?

Megan Bergkessel

University of Dundee

Regulation of protein synthesis during growth arrest

Abstract:

Growth arrest due to nutrient limitation is a very common state for bacteria in a wide range of natural environments, whether living as symbionts or pathogens of multicellular creatures or members of diverse microbial communities. We have used tools such as Bio-Orthogonal Non-Canonical Amino acid Tagging (BONCAT), fluorescence in-situ hybridisation, and reporter strains to measure biosynthesis during such nutrient-limited growth arrest at the level of individual cells. We are combining this information with population-level measurements of protein and RNA abundance for specific putative regulators. Ultimately, we seek to uncover regulatory pathways that govern gene expression during growth arrest and understand how they shape temporal and population-level heterogeneity in protein synthesis rates. We have surveyed patterns of starvation survival and ongoing protein synthesis during carbon starvation for several different Proteobacteria. Currently, we are working to identify and characterise regulators that support the low levels of ongoing protein synthesis observed during starvation in Pseudomonas aeruginosa. Even though average protein synthesis rates during growth arrest are very low, we observe heterogeneity within populations, species-specific differences, and complex responses to environmental changes."

List of speakers, continued (alphabetical)

Michael A. Sørensen
University of Copenhagen
The degradation rate of ribosomes during stationary phase depends on the type of missing nutrient.

Morten Nørholm
Technical University of Denmark
Molecular mechanisms in the evolution of nutrient-limited bacterial colonies
Abstract:
The evolution of microorganisms often involves changes of unclear relevance, such as transient phenotypes and sequential development of multiple adaptive mutations in hotspot genes. Previously, we showed that ageing colonies of an *E. coli* mutant unable to produce cAMP, when grown on maltose, accumulated mutations in the crp gene (encoding a global transcription factor) and in genes involved in pyrimidine metabolism such as cmk; combined mutations in both crp and cmk enabled fermentation of maltose (which usually requires cAMP-mediated CRP activation for catabolic pathway expression). Here, we study the sequential generation of hotspot mutations in those genes, and uncover a regulatory role for pyrimidine nucleosides in carbon catabolism. Cytidine binds to the cytidine regulator CytR, modifies the expression of sigma factor 32 (RpoH), and thereby impacts global gene expression. In addition, cytidine binds and activates a Crp mutant directly, thus modulating catabolic pathway expression, and could be the catabolite modulating factor whose existence was suggested by Jacques Monod and colleagues in 1976. Therefore, transcription factor Crp appears to work in concert with CytR and RpoH, serving a dual role in sensing both carbon availability and metabolic flux towards DNA and RNA. Our findings show how certain alterations in metabolite concentrations (associated with colony ageing and/or due to mutations in metabolic or regulatory genes) can drive the evolution in non-growing cells.

Nathalie Balaban
Hebrew University of Jerusalem
The Impact of the Disrupted Cellular State on Antibiotic Response

Oleg Igoshin
Rice University
Making cell-fate-decisions: lessons from *B. subtilis*
Abstract:
I will describe how theoretical predictions and their experimental tests uncover how model bacteria Bacillus subtilis decides between sporulation and biofilm formation cell-fates. We show how these can detect starvation stress without detecting any specific metabolites by sensing the growth rate and associated changes in gene dosage of key genes."

Petra Levin
Washington University
The environment shapes the cell

Rami Pugatch
Ben-Gurion University of the Negev
A unifying autocatalytic network-based framework for bacterial growth laws.
Abstract:
Bacterial cells contain various autocatalytic cycles, e.g., the well-known ribosome cycle, where ribosomes translate ribosomal proteins that subsequently self-assemble to form new ribosomes. In this talk I will explain how the transcription-translation machinery couples all cellular autocatalytic cycles, resulting in balanced exponential growth. I will explain how all known growth laws stem from this unifying picture, without invoking optimality. I will present the RNA polymerase (RNAP) growth law based on the RNAP autocatalytic cycle, where RNAPs transcribe mRNAs of its constituent Rpo protein subunits. Before degrading, these mRNAs catalyze Rpo proteins employing ribosomes. The Rpo proteins subsequently self-assemble, forming new RNAPs, thus completing the cycle. Contrary to ribosome growth law, a reduction in growth rate due to a shortage in RNAPs occurs without affecting the ribosomal protein mass fraction."

List of speakers, continued (alphabetical)

Roberto Kolter
Harvard Medical School
Thoughts on the Physiology of Natural Bacterial Populations

Seungeon Oh
Harvard Medical School
Quantitative single-cell physiology of growth and size regulation using label-free microscopy

Severin Schink
Harvard Medical School
Quantitative insights into bacterial starvation survival
Abstract:
The majority of microbes on earth, whether they live in the ocean, the soil or in animals, are not growing, but instead struggling to survive starvation. Some genes and environmental conditions affecting starvation survival have been identified, but despite almost a century of study, we do not know which processes lead to irreversible loss of viability, which maintenance processes counteract them and how lifespan is determined from the balance of these opposing processes. In my talk, I will present time-lapse microscopy that captures the cell death process of *E. coli* during carbon starvation for the first time. We found that a lack of nutrients results in the collapse of ion homeostasis, triggering a positive-feedback cascade of osmotic swelling and membrane permeabilization that ultimately results in lysis. Based on these findings, we hypothesized that ion transport is the major energetic requirement for starving cells and the primary determinant of the timing of lysis. We therefore developed a mathematical model that integrates ion homeostasis and 'cannibalistic' nutrient recycling from perished cells to predict lifespan changes under diverse conditions, such as changes of cell size, medium composition, and prior growth conditions. Guided by model predictions, we found that cell death during starvation could be dramatically slowed by replacing inorganic ions from the medium with a non-permeating osmoprotectant, removing the cost of ion homeostasis and preventing lysis. Our quantitative and predictive model explains how survival kinetics are determined in starvation and elucidates the mechanistic underpinnings of starvation survival.

Shiladitya Banerjee
Carnegie Mellon University
Cell morphology as regulator of bacterial fitness and adaptation

Steen Pedersen
University of Copenhagen
The genetic code, in addition to encode the amino acids, also encodes signals that modify gene expression by affecting mRNA half-life and premature transcription termination.

Stefan Klumpp
University of Göttingen
Resource allocation in bacterial motility

Stephen Cooper
University of Michigan Medical School
Three Critical Manifestos for the Cell Cycle: On Synchronization, on G0, on Gene Expression

List of speakers, continued (alphabetical)

Susan Gottesman
National Cancer Institute, NIH
Coming and Going: *E. coli* circuitry for adjusting to and recovering from stress

Sven van Teeffelen
University of Montreal
Regulation of mass density in rod-shaped bacteria
Abstract:
Bacteria such as the rod-like *Escherichia coli* control intracellular density remarkably well during steady-state growth. My lab identified and measured two major variables responsible for mass density that are seemingly independently controlled on the generation timescale: First, cells control the surface-area-to-mass ratio by coupling the rate of global surface-area growth to biomass growth. Interestingly, the rate of surface growth is independent of cell-wall insertion in *E. coli* and only partially dependent on cell-wall insertion in the Gram-positive Bacillus subtilis.
Second, cells control cell width, which is inversely proportional to the surface-to-volume ratio. On long time scales width regulation and surface-to-mass ratio are correlated, which contributes to partial density homeostasis. We show that osmotic pressure (turgor) has an important role in this process.

Søren Molin
Technical University of Denmark
Investigations of microbial physiology in adapting bacterial populations involved in airway infections

Tanneke den Blaauwen
University of Amsterdam
Quantitative analysis of the timing of midcell localization of division proteins reveals specific roles for endopeptidases PBP4 and PBP7

Terence Hwa
UC San Diego
Sculpting of the gene expression landscape by physiological constraints

Teuta Pilizota
University of Edinburgh
The proton motive force determines *Escherichia coli*'s robustness to extracellular pH
Abstract:
Maintaining intracellular homeostases is a hallmark of life, and key physiological variables, such as cytoplasmic pH, osmotic pressure, and proton motive force (PMF), are typically interdependent. Developing a mathematical model focused on these links, we predict that *Escherichia coli* uses proton-ion antiporters to generate an out-of-equilibrium plasma membrane potential and so maintain the PMF at the constant levels observed. The strength of the PMF consequently determines the range of extracellular pH over which the cell is able to preserve its near neutral cytoplasmic pH. In support, we concurrently measure the PMF and cytoplasmic pH in single cells and demonstrate both that decreasing the PMF's strength impairs *E. coli*'s ability to maintain its pH and that artificially collapsing the PMF destroys the out-of-equilibrium plasma membrane potential. We further predict the observed ranges of extracellular pH for which three of *E. coli*'s antiporters are expressed, through defining their cost by the rate at which they divert imported protons from generating ATP. Taken together, our results suggest a new perspective on bacterial electrophysiology, where cells regulate the plasma membrane potential by changing the activities of antiporters to maintain both the PMF and cytoplasmic pH.

List of speakers, continued (alphabetical)

Thomas Julou
University of Basel
Growth-rate controls the sensitivity of gene regulatory circuits.
Abstract:
Unicellular organisms adapt to their changing environments by gene regulatory switches that sense chemical cues and induce specific target genes when the inducing signal is over a critical threshold. We here demonstrate that the response of these switches is highly context-dependent; more specifically, their sensitivity decreases with growth rate due to a natural competition between uptake of the inducer and dilution due to growth. Mathematical modelling reveals that this growth-coupled sensitivity is a general feature of regulatory switches, independent of their architecture. We show experimentally that the concentration of inducer required for activating the lac operon in *E. coli* critically depends on growth rate, both at the single-cell and at the population level. Moreover, we establish that growth-coupled sensitivity allows bacteria to implement a concentration-dependent sugar preference, in which a new carbon source is used only if its concentration is high enough to increase growth rate. We demonstrate experimentally that this strategy governs how mixtures of glucose and lactose are used, and is implemented through the central regulator CRP. Overall growth-coupled sensitivity provides a general mechanism through which cells can 'mute' external signals in beneficial conditions when growth is fast, and become highly sensitive to alternative nutrients or stresses when growth is slow or arrested."

Tsutomu Katayama
Kyushu University
Dynamic mechanisms constituting regulation for the replication initiator DnaA protein in *Escherichia coli*

Vahid Shahrezaei
Imperial College London
Mechanisms of size control across growth conditions in *E coli* and Pombe

Willie Donachie
University of Edinburgh
Answering the Nordström Question (and maybe a few others).

Yuichi Wakamoto
University of Tokyo
Raman-omics correspondence reveals homeostatic core and stoichiometrically-balanced dynamical groups inside cells

Yusuke Himeoka
University of Tokyo
Emergence of growth and dormancy from a kinetic model of the *Escherichia coli* central carbon metabolism

List of posters

- All posters will be up for the duration of the meeting.
- Poster presenters with even numbers present on Monday, June 13
- Poster presenters with uneven numbers present on Tuesday, June 14

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Ala Trusina
University of Copenhagen

2

Alba García Vázquez
University of Copenhagen

PLANET EARTH-LIKE PATTERNS IN THREE-DIMENSIONAL MULTI-SPECIES BACTERIAL COLONIES.

Abstract:
Bacteria typically grow in communities and this provides them substantial advantages compared to solitary cells. These communities are often comprised of multiple bacterial species leading to the emergence of complex spatial patterns. The emergence of these complex spatial patterns can have a profound effect on bacterial function and survival within the communities. Most experimental studies investigating the mechanism behind this pattern formation have focused in two-dimensional systems. Here we propose a novel approach to study three-dimensional multi-species bacterial colonies. This three-dimensional setting replicates better some environmental bacterial habitats such as soil and intestines. Our results indicate that in three-dimensional multi-species colonies just the cells from the outer part of the colony are able to grow while the center of the colony remains static. We anticipate our protocol to be a starting point for further studies. For example, the protocol could be used to bring some light to many different biological and physical questions which are still unanswered such as the mechanism behind horizontal gene transfer and the social interactions arising within multi-species three-dimensional bacterial colonies. Furthermore, understanding how bacteria thrive in competitive habitats and their cooperative strategies for surviving extreme stress can be instructive, for instance, to inspire new investigations for developing a more rational approach for battling pathogenic bacteria resistant to antibiotics.

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Alfonso Soler Bistue
Universidad Nacional de San Martín / Consejo Nacional de Investigaciones Científicas y Técnicas (UNSAM/CONICET)

4

Andreas Berglund Nielsen
University of Copenhagen

5

Anna Knöppel
ICM, Uppsala University

The coordination of replication initiation to growth rate in Escherichia coli

List of posters, continued.

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Ardré Maxime
ESPCI

High throughput millifluidic reveals that bacterial population of Pseudomonas fluorescens follows a cell leader to exit its lag phase.

Abstract:
Maxime Ardré(a), Guilhem Douclier(a), Naama Brenner(b), Paul Rainey(a,c)
a) Laboratoire Biophysique et Évolution, CBI, ESPCI Paris, Université PSL, CNRS 75005 Paris, France
b) Network Biology Research Laboratories, and Department of Chemical Engineering, Technion–Israel Institute of Technology, Haifa, Israel
c) Department of Microbial Population Biology, Max Planck Institute for Evolutionary Biology, Plön, Germany

The relationship between the number of cells colonizing a new environment and time for resumption of growth is a subject of long-standing interest. In microbiology this is known as the “inoculum effect”. Its mechanistic basis is unclear with possible explanations ranging from the independent actions of individual cells, to collective actions of populations of cells. Progress requires precise measurement of lag-time distributions while at the same time, experimentally controlling inoculum size. Here we use a millifluidic droplet device in which the growth dynamics of hundreds of populations founded by different numbers of Pseudomonas fluorescens cells, ranging from a single cell, to one thousand cells, were followed in real time. Our data show that lag phase decreases with inoculum size. The average decrease, variance across droplets, and distribution shapes, follow predictions of extreme value theory, where the inoculum lag-time is determined by the minimum value sampled from the single-cell distribution. Our experimental results show that exit from lag phase depends on strong interactions among cells, consistent with a ""leader-cell"" triggering end of lag phase for the entire population.
Exit from lag phase in populations of Pseudomonas fluorescens is determined by strong interactions among cells,
Maxime Ardré, Guilhem Doulcier, Naama Brenner, Paul B. Rainey
bioRxiv 2022.01.24.477561; doi: https://doi.org/10.1101/2022.01.24.477561"

7

Benjamin Roller
University of Vienna

Single-cell mass accumulation for vibrios experiencing nutrient fluctuations

Abstract:
Nutrient availability fluctuations are a common ecological challenge for bacteria. Microbial physiologists have developed a quantitative understanding of how bacteria change their size and composition when nutrients fluctuate. However, building from physiological principles to a quantitative understanding of how bacteria grow in nature requires a reconciliation between single-cell and population growth phenomena. In this study, we are quantifying mass accumulation during a variety of nutrient fluctuations for individual cells and populations of Vibrio cyclitrophicus isolated following an algal bloom. We are developing multiple modalities for measuring the mass of individual bacteria using a suspended microchannel resonator. With these sensors we can perform either high-throughput snapshots of single-cell mass distributions of populations or high-resolution mass accumulation time course measurements of individual cells. We find that cells overshoot their optimal steady-state mass when recovering from standard overnight culture starvation in batch culture before divisions return cell mass slowly back to its steady-state value. With these techniques we are beginning to measure single-cell and population growth physiology in the discontinuous, heterogeneous nutrient landscape these bacteria face in nature. These data address the existing conceptual and technological gaps between quantitative microbial physiology and how bacteria grow in the wild."

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Cristina Amador
University of Copenhagen

List of posters, continued.

9

Cristina Palma
Tampere University

Changes in DNA supercoiling contribute to the triggering of the short-term, cold shock transcriptional program of E. coli

Abstract:
Cristina S.D. Palma, Suchintak Dash, Ines S.C. Baptista, Mohamed N.M. Bahrudeen, Bilena L.B. Almeida, Vatsala Chauhan, Rahul Jagadeesan, and Andre S. Ribeiro
Laboratory of Biosystem Dynamics, Faculty of Medicine and Health Technology, Tampere University, Arvo Ylpön katu 34, 33520, Finland. E-mails: cristina.santosdiaspalma@tuni.fi; andre.sanchesribeiro@tuni.fi.
Present techniques have allowed identifying genes responsive to temperature shifts, including cold shock. Yet, the evolved mechanics that make them responsive remains elusive. We hypothesized that, for short-term responsive genes, their mechanisms are intrinsic, physics-based, and present in many, but not all genes (or at least, not sufficiently strong in all genes to influence them). Further, for those genes repressed in cold shock, their mechanisms ought to be able to override any activation mechanisms. Thus, transcription locking due to the reduction of negative supercoiling levels could be the mechanism triggering short-term, cold shock repressed (CSR) genes. In support, DNA supercoiling influences genome-wide transcription levels, decreasing negative supercoiling enhances transcription locking, and the unlocking process is energy-dependent. Here, we identified short-term, cold shock repressed (CSR) genes, as well as supercoiling sensitive (SS) genes by RNA-seq. Interestingly, many CSR genes are also SS, and the response strengths to CS and to Gyrase inhibition are correlated. Next, having ruled out transcription factors and global regulators as being influential and shown that the single-cell variability in protein numbers increases with CS, we present findings on biophysical phenomena supporting our hypothesis. In the end, we propose a transcription model of how supercoiling sensitivity generates short-term CSR genes. Our results may assist in presetting the temperature sensitivity of future synthetic circuits, e.g., potentially by carefully selecting their chromosome location."

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Ditte Heidemann
University of Copenhagen

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Elin Larsson
California Institute of Technology

A ratiometric reporter for phosphorus limitation in a wheat-colonizing Pseudomonad

Abstract:
Phosphorus is an element required by all life. In addition to nitrogen and potassium, it is important for growing crops and is added to soil in fertilizers. Unlike nitrogen, however, phosphorus is not a renewable resource as it has no closed biogeochemical cycle. Further, overfertilization leads to runoff that causes increased emissions of greenhouse gases and loss of biodiversity in aquatic ecosystems. Determining the amount of phosphorus fertilization that a soil needs will require a quantitative, dynamic measure of bioavailable phosphorus. Towards this end, microbes may be able to help, both by detecting bioavailable phosphorus and being able to recycle it. Whether bioengineering is done for the purpose of biosensing or actuation, it is important to use organisms other than E. coli for designing field deployable microbes that are better adapted to the soil environment, thereby enabling long-term survival. Pseudomonas synxantha is a Gram-negative bacterium that lives in the Dryland wheat rhizosphere of the Pacific Northwest. In return for being fed carbon by the wheat root exudates, it has biocontrol properties that protect the wheat from disease. We are working with this species to develop a genomically integrated phosphorus bioreporter system that couples fluorescence detection to production of an electronic signal output. Here, we design, build, and integrate a ratiometric reporter in P. synxantha and characterize its response at different phosphate concentrations, other nutrient limitations and at cell densities where quorum sensing is at play.

List of posters, continued.

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Ferdinand Greiss
Weizmann Institute of Science

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Glen Dsouza
ETH-Zurich

14
Itzhak Fishov
Ben-Gurion University of the Negev
Phenotypic variability in *E. coli*: Stochastic nucleoid segregation dynamics as a possible source
Abstract:
Bacteria display broad phenotypic variability in normal growth conditions, employing it as an efficient strategy to deal with external stress. Phenotypic variability in bacterial cells is currently ascribed to both the gene expression noise and the nonrandom partitioning of low copy number components. One of the central processes in the bacterial cell cycle is the separation of genetic material between daughter cells. The chromosome segregation dynamics appears as sequential, progressing along with the replication process, but highly variable in its rate and the resulting nucleoid localization within the cell. The chromosome segregation variability may, on one hand, reflect the flexibility necessary for correlation with other major cellular processes and, on the other hand, represent an inherent source of bacterial cell phenotypic variability. We followed the segregation dynamics of nucleoids in normally dividing lineages of Escherichia coli and non-dividing filaments, in which the heterogeneity produced by gene expression noise and uneven partitioning of freely diffusing proteins is minimized. Quantitative characteristics of this dynamics reveal that the mean partitioning rate of nucleoids is essentially the same in dividing and filamentous cells, displaying a substantial variability in both. However, the absence of caps separating the cells improves the synchrony of nucleoid separation and reduces variability in distance between them. The remaining variability is inherent to the nucleoid hyperstructure. Our results also suggest that the nucleoid segregation mechanism is only weakly dependent on cell division.

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Jakob Frimodt-Møller
University of Copenhagen

The role of rpsLI82N in resistance to an antimicrobial antisense agent acting on mRNA

Abstract:
Jakob Frimodt-Møller[1], Irene R. Martinez[1], Thomas Prossliner[1], Lotta J. Happonen[2], Peter E. Nielsen[3], and Anders Løbner-Olesen[1]

1. Department of Biology, University of Copenhagen, Denmark
2. BioMS, Lund University, Sweden
3. Department of Cellular and Molecular Medicine, University of Copenhagen, Denmark

Antimicrobial antisense agents inhibit gene expression at the translational level via specific binding to sequence complementary mRNA of essential genes or to essential sites of rRNA, leading to growth cessation. Due to their biological stability, neutral charge, high binding affinity and specificity for sequence complementary RNA, synthetic nucleic acid analogs such as peptide nucleic acids (PNA) have been proposed as a future antibiotic. Unless stated otherwise, the PNA part always targets translation of the acpP mRNA encoding the acyl carrier protein, which is essential for fatty acid synthesis. Naked PNA are unable to traverse the cell envelope of bacteria. Hence, these requires a carrier molecule, e.g. bacterial penetrating peptide (BPP), for efficient translocation into the bacterial cytoplasm. In preliminary studies, adaptive laboratory evolution were preformed to obtain peptide-PNA resistant mutants.

Here, all isolated BPP-PNA resistant mutants contained a single missense mutation in rpsL (rpsLI82N). rpsL encodes for the S12 protein that is involved in the inspection of codon–anticodon pairings in the ribosomal A site, as well as in the succeeding domain rearrangements of the 30S subunit that are essential for accommodation of aminoacyl-tRNA. Apart from the rpsLI82N mutation, other S12 mutants that do not affect the sensitivity to streptomycin have been isolated. Testing a sub-set of these surprisingly showed that other rpsL mutations also conferred resistance to BPP-PNA. These along with rpsLI82N will be referred to as rpsL*. Thus, either these point-mutations all results in a specific S12 configuration that confers resistance to BPP-PNA or multiple different configurations of S12 can do the same. Here, I will go in-depth with these newly identified rpsL mutations to understand how a mutated ribosome can circumvent PNA binding and translate mRNA, if the mutated ribosome can “displace” other regulatory molecules acting at the mRNA level, and their effect on cellular physiology.“

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Juanita Lara-Gutierrez
ETH Zürich

List of posters, continued.

17

Jumpei Yamagishi
The University of Tokyo

Microeconomics of Metabolism

Abstract:
Metabolic behaviors of proliferating cells are often explained as a consequence of rational optimization of cellular growth rate, whereas microeconomics formulates consumption behaviors as optimization problems. Here, we pushed beyond the analogy to precisely map metabolism onto the theory of consumer choice. We thereby revealed the correspondence between long-standing mysteries in both fields: the Warburg effect, a seemingly wasteful but ubiquitous strategy where cells favor aerobic glycolysis over more energetically efficient oxidative phosphorylation, and Giffen behavior, the unexpected consumer behavior where a good is demanded more as its price rises. We identified the minimal, universal requirements for the Warburg effect: a trade-off between oxidative phosphorylation and aerobic glycolysis and complementarity, i.e. impossibility of substitution for different metabolites. Thus, various hypotheses for the Warburg effect are integrated into an identical optimization problem with the same universal structure. Besides, the correspondence between the Warburg effect and Giffen behavior implies that oxidative phosphorylation is counter-intuitively stimulated when its efficiency is decreased by metabolic perturbations such as drug administration or mitochondrial dysfunction; the concept of Giffen behavior bridges the Warburg effect and the reverse Warburg effect. The possibility of application of microeconomics to general metabolic systems will be also discussed.

List of posters, continued.

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Ken-ichiro Kamei
The University of Tokyo

Stoichiometry-Based Low-Dimensional Structure of Proteome Revealed by Raman-Proteome Correspondence

Abstract

To understand the dynamical principles driving biological cells, an appropriate coarse-grained picture derived from the comprehensive cellular components' dynamics is needed. Vibrational spectroscopy techniques such as Raman spectroscopy potentially provide the information of comprehensive molecular composition of cells. Unlike existing omics technologies, Raman spectroscopy has the advantage that it can be applied to living cells. Although biological interpretation had been assumed difficult or even impossible due to their complexity, it was previously shown that cellular Raman spectra could be linked to cellular transcriptomes (Kobayashi-Kirschvink et al., 2018); changes in Raman spectra might be therefore interpretable through the scope of omics profiles.

Despite its promising potential for cell analysis, it remains unclear whether cellular Raman spectra can also be linked to other layers of omics such as proteomes and why such Raman-omics linkages can be found. To address these questions, we cultured E. coli under the conditions used for obtaining the large-scale quantitative E. coli proteome data (Schmidt et al., 2016) and measured Raman spectra of the cells. First, we found that the Raman spectra and the proteomes are related approximately linearly and that the global changes of proteomes are predictable from the Raman spectra. Furthermore, by dissecting the coefficients of the obtained linear relation, we found a group of proteins that conserved their abundance ratios across the conditions, which we named "homeostatic core". This group contained many molecules involved in processing genetic information and was directly related to "bacterial growth law" (Scott et al., 2010). Next, we devised a method to extract the groups of proteins that maintain their stoichiometry without relying on Raman data. We thereby determined the "homeostatic core" as well as several groups of proteins expressed coordinately under specific conditions only from omics data. To further delve into the proteome constraints and the mechanism of the Raman-proteome correspondence, we visualized the low-dimensional proteome structures in two ways, firstly using the Raman-proteome conversion coefficients and secondly based on the Laplacian eigenmaps of the stoichiometry conservation strength matrix. Surprisingly, we found that the distributions of the stoichiometry-balanced groups in these distinct low-dimensional subspaces are similar, with the homeostatic core located at its center and the condition-specific groups at its periphery. We analyzed mathematically the relation between these two subspaces and the implication of the characterized similarity, revealing two crucial points: (i) Axes distinguishing condition-dependent differences of Raman spectra nearly correspond to axes explaining stoichiometry-based module structure of proteomes, and (ii) a global constraint exists between expression patterns across conditions and stoichiometric balance of omics components. Lastly, we confirmed such similarity between Raman and omics low-dimensional structures and the global constraints on omics dynamics even for other organisms and perturbation. Our results indicate that changes of complex cellular Raman spectra reflect the stoichiometric balance of omics components, and that stoichiometric balance and dynamics of abundance ratios of cellular components are fundamental to the cellular system."

List of posters, continued.

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Laura Martínez Alvarez
University of Copenhagen

To be or not to be an anti-CRISPR protein: SIRV2gp37 and the importance of working with native biological systems

Abstract:
Viral members of the protein family DUF1874 have been reported to act as anti-CRISPR (acr) proteins that degrade cyclic tetra-adenylate (cA4), a nucleotide secondary messenger produced after the activation of several type III CRISPR-Cas systems in bacteria and archaea. Specifically, protein SIRV1gp29 was shown to inhibit type III-A and type III-B CRISPR systems in plasmid-born assays in heterologous systems. In this work we investigate the function of SIRV2gp37, a close homolog of SIRV1gp29.
We demonstrate that GP37 has no anti-CRISPR activity during infection of Sulfolobus islandicus LAL14/1 with SIRV2, although it is able to protect SIRV2 from type III targeting when GP37 is expressed from a plasmid. The inability of GP37 to act as an acr in the native, biological system is due to the protein expression timing: GP37 is a middle-late gene, thus unable to inhibit CRISPR-Cas targeting at the onset of infection. We find that while GP37 is a non-essential gene, it confers a mild replicative advantage to the virus. This advantage is mediated, in hosts with active CRISPR-Cas targeting, by the interaction between GP37 and host protein SiL1451, which results in the inhibition of the protein lysine methyltransferase activity of SiL1451, responsible for extensive methylation of surface lysines of two-thirds of the cellular proteins.
Plasmid-based experiments have allowed the discovery and characterization of tenths of prokaryotic defense systems in the recent years. Although this experimental strategy has several advantages, our study highlights the importance of working with biological systems as close as possible to the native conditions, and the limitations of extrapolating results obtained using heterologous systems."

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Michael Manhart
ETH Zurich

Physiology, growth, and evolution of microbes under simultaneous limitation for multiple nutrients

Abstract:
Microbial growth relies on the presence of several nutrients, including elemental nutrients such as carbon and nitrogen, as well as complex nutrients like vitamins and amino acids. Evidence from biogeochemistry, especially in aquatic environments, suggests that multiple nutrients may be simultaneously rare in nature and therefore limit growth. However, we poorly understand how this co-limitation affects microbial physiology, growth, and evolution. We introduce a framework for quantifying nutrient limitation of both growth rate and biomass production based on metabolic control analysis. We first show that the degree of co-limitation between two nutrients corresponds to the structure of the metabolic pathway integrating the nutrients. We then show that, depending on the metabolic structure, co-limitation of those nutrients can spontaneously emerge as nutrient affinities evolve to match environmental availability. Finally, we experimentally measure growth across a range of co-limiting nutrient pairs to assess their effects on the proteome and growth phenotypes. Altogether our results suggest that nutrient co-limitation, rather than single limitation, is a generic property of microbial populations."

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Mikkel Svenningsen
University of Copenhagen

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Mireia Cordero
University of Copenhagen

List of posters, continued.

23

Mogens Kilstrup
Technical University of Denmark (DTU)

Prequisites for a purely stochastic phage switch model, capable of taking lysis/lysogeny decisions

Abstract

Johan Cassias and Mogens Kilstrup*

Phage switches are usually bi-stable as most genetic switches. In addition, phage switches should be capable of taking the initial decision between the two switch states. To analyze this important function of a phage switch, we have focused on a specific switch family whose members have been found in genetically unrelated phages like Staphycoccus phages 13 and PVL, and Lactococcus phages TP901-1 and Tuc2009. The switch is contained on an 1100 to 1300 bp DNA fragment, which encodes only two regulators, a CI repressor and a modulator of repression (Mor). We have designed a stochastic non-equilibrium (Gillespie) model, with reactions between i) RNA polymerase and the PL and PR promotes including closed and open complexes, ii) between CI and OR, OL, and OD operator sites, iii) between MOR and CI, and iv) between the CI:MOR complex and OM. Simulations in cell volume of 1.7 fL was also performed with different transcription time for the long cI (30 sec) and short mor (15 sec) genes, as well as cooperativity in CI binding. A benefit from the simulation in a volume of 1.7 10-15 L is that one molecule in this volume is equal to 1 nM, so that all experimental rate constants given in M could just be multiplied by 10-9 nM/M and thereby be normalized to nM.

The design of the simulation model started by simulating expression from a single promoter, which we based upon the kinetic constants for the T7A1 promoter published by Bianca Sclavi et al. We show that this promoter follows a Michaelis Menten type saturation kinetics, with a KM around 55 nM free RNA polymerases. At three different RNApol concentrations, we then analyzed the sensitivity of the gene expression level from the promoter if all rate constants were varied from 1/100-fold to 100-fold. Subsequently CI repression, MOR anti-repression, CI:MOR repression, and all other factors were gradually introduced.

In our analysis of the decision outcome space, we had envisioned that our stochastic model would lead to a clean separation of events with either high or low CI/MOR ratio. However, our simulations showed that a prerequisite of a small percentage of lysogenic decisions posed the restriction that the maximal CI expression rate should be much smaller than the maximal MOR expression rate. For CI to win at such low odds, high-CI/MOR decisions required a reinforcement step. The few lucky situations in which early CI expression was fast enough to get ahead of MOR expression, had to result in cooperative binding to OL, OR, and OD through CI-hexamer formation.

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Mohammad Roghanian
Rigshospitalet

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Muriel Schicketanz
University of Copenhagen

Molecular Regulation of SpoT in *E. coli*

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Ole Skovgaard
Roskilde University

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Pranas Grigaitis
Vrije Universiteit Amsterdam

Optimal resource allocation explains energetics-related costs of metabolic strategies in budding yeast

Abstract:
Optimal allocation of available cellular resources is central for microorganisms to strive and outcompete individuals of the same and other microbial species. Allocation of resources mainly manifests as production of proteins required to sustain growth, and thus shapes the metabolic strategies microbes undertake. In different conditions, different metabolic strategies are superior to others (sustain fastest growth per unit protein), and will be adopted as a result of optimal resource allocation. One of the main assets for growth is energy, and thus cells invest generously into proteins, associated with energy harvesting - up to 1/3 of the total proteome of budding yeast Saccharomyces cerevisiae, the organism studied here. However, the nature of energetic costs in different environments remains poorly characterized, thus here we aimed to gain insight into condition-specific energetic costs of S. cerevisiae growth.
We constructed an updated version the proteome-constrained (pc-) model of S. cerevisiae, pcYeast8. We used the pcYeast8 model in order to characterize condition-dependent metabolic strategies: we simulated growth in different nutrient-limited conditions, and predicted metabolic fluxes and protein abundance. Moreover, we performed simulations where we perturbed the proteome capacity of different compartments. We suggest that additional growth-associated energetic costs are present in anaerobic environments, and we have identified the mitochondrial capacity as the constraint regulated by the expression level of the transcription factor Hap4. We conclude that the fine-grained modeling of resource allocation in S. cerevisiae gives new insights into the role of energy harvesting to its physiology.“

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Ralf Steuer
Humboldt-University of Berlin

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Rocio Espinosa
University of Copenhagen

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Susanne Häussler
Rigshospitalet

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Thomas Søndergaard Stenum
Uppsala University

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Yong Everett Zhang
University of Copenhagen