BP 18: Focus Session: Physics of Microbial Systems - organized by Tobias Bollenbach and Benedikt Sabass

Time: Wednesday 9:30–13:00 Location: H 2013

Invited Talk BP 18.1 Wed 9:30 H 2013

Dynamics of cellular metabolism, size, and motility — •Sander Tans — AMOLF, Amsterdam, the Netherlands

We use time-lapse microscopy to measure the dynamics of individual cells, focusing on a number of different questions. I will present work on the relation between fluctuations in the expression of catabolically active enzymes and cellular growth, how cells control their size in the presence of external and internal perturbations, and a surprising observation of motility in epithelial cells that is triggered by viral infection.

BP 18.2 Wed 10:00 H 2013

Salmonella **Typhimurium in the search of host cells** — Emiliano Perez Ipiña[1], Stefan Otte[1], Rodolphe Pontier-Bres[2], Dorota Czerucka[2], and •Fernando Peruani[1] — [1]Université Côte d'Azur, Laboratoire J.A. Dieudonné, UMR 7351 CNRS, Nice, France — [2]Centre Scientifique de Monaco (CSM), Principality of Monaco

Combining experiments and theory, we study how *Salmonella* Typhimurium (ST) search for human T84 colonic epithelial cells (HC), which, anchored on the bottom surface of a chamber, are invaded by ST. Our study reveals that near the surface ST do not display biased motion towards HC and the localization of HC involves a random search. We find that this random search has a well-defined average search time $\langle\tau\rangle$, which is determined by the details of the near-surface motion of ST and particularly by the diffusion coefficient D. We show that this random search can be well-described by a model, analytically tractable, of chiral active particles with active speed fluctuations and find that these fluctuations are of biological origin and account for up to 40% of D. Using simple arguments and simulations, we show that the number of ST that invade HC (NIB) is fully determined by $\langle\tau\rangle$, proving that D controls $\langle\tau\rangle$, and $\langle\tau\rangle$ determines NIB. Furthermore, our study reveals that within the same bacterial population (same genome), there exists a large range of inter-individual variability of the bacterial exploring capacity, with D ranging over four orders of magnitude. This finding together with the relation between D, $\langle\tau\rangle$, and NIB suggests that the individual infection capacity is highly heterogeneous within the same bacterial population.

BP 18.3 Wed 10:15 H 2013

Sex or Simplicity: Phenotypic interference and the cost of complexity in asexual evolution — Torsten Held[1,2], •Daniel Klemmer[1,2], and Michael Lässig[1] — [1]Institut für Theoretische Physik, Universität zu Köln, Köln, Deutschland — [2]Equal contribution

The asexual evolution of microbes and viruses often generates clonal interference, a mode of competition between genetic clades within a population. We show that interference strongly constrains genetic and phenotypic complexity. Our analysis is based on a minimal biophysical model that represents each gene by a quantitative molecular phenotype, its fold stability. The model displays a generic mode of asexual evolution called phenotypic interference, which occurs over a wide range of evolutionary parameters appropriate for microbial populations. It generates a strong burden of complexity: The fitness cost of mutations increases faster than linearly with the number of genes. We show that recombination eliminates the superlinear cost through a first-order phase transition to a mode of sexual evolution. This implies a large fitness advantage of even facultative recombination and provides a biophysically grounded scenario for the evolution of sex. In a broader context, our analysis suggests that the systems biology of microbial organisms is strongly intertwined with their mode of evolution.

BP 18.4 Wed 10:30 H 2013

Dormant, dead or alive: measuring steady state free energy levels in bacterial cells — •Leonardo Mancini and Teuta Pilizota — University of Edinburgh

Bacteria can survive a variety of external stresses by entering a state of suspended growth that is commonly referred to as dormancy. Such response has historically been considered a unequivocal low metabolism-low energy state and a vast array of stressors seem to be avoidable through dormancy. Antibiotics are among the most notable examples of such stressors and tolerant, dormant cells are known as persisters. However, recent experiments show that some persisters might survive antibiotic challenges through mechanisms that are, in contrast, energy consuming. The findings open the possibility of several different dormant steady states with distinct cellular free energy levels. To verify such a hypothesis, molecular sensors that can provide information on cellular energetics in vivo and at the single cell level are needed. To this end, we have successfully optimized the expression of a previously reported QUEEN ATP sensor and characterized in E. coli the newly proposed membrane voltage dye, Thioflavin T. Our results provided insights that can be generalized to other dyes, such as TMRM and DiSC3(5). Using the sensors, we present measurements of free energy levels during dormancy when this is induced by different conditions and signals, such as starvation, quorum sensing, and stress signalling molecules.

BP 18.5 Wed 10:45 H 2013

Localized hypermutations govern competition dynamics through positioning in bacterial colonies — •Robert Zöllner, Enno Oldewurtel, Nadzeya Kouzel, and Berenike Maier — Department of Physics, University of Cologne, Zülpicher Str. 77, 50539 Köln, Germany

Cellular positioning towards the surface of bacterial colonies and biofilms can enhance dispersal, provide a selective advantage due to increased nutrient and space availability, or shield interior cells from external stresses. Little is known about the molecular mechanisms that govern bacterial positioning. Using the type IV pilus (T4P) of Neisseria gonorrhoeae, we tested the hypothesis that localized hypermutations govern positioning and thus enhance bacterial fitness in expanding gonococcal colonies. By independently tuning growth rate and T4P-mediated interaction forces, we show that the loss of T4P and the subsequent segregation to the front confers a strong selective advantage. Sequencing of the major pilin gene of the spatially segregated sub-populations and an investigation of the spatio-temporal population dynamics was carried out. Our findings indicate that localized hypermutations generate a standing variation of pilin sequences within the inoculation zone, while variants associated with a non-piliated phenotype segregate to the front of the growing colony. We conclude that tuning of attractive forces by mutations is a powerful mechanism for governing the population dynamics of bacterial colonies.

15 min. break

BP 18.6 Wed 11:15 H 2013

Quantitative modeling of nutrient-limited growth of bacterial colonies in microfluidic cultivation — •Jens Elgeti — Theoretical Soft Matter and Biophysics, ICS-2, Forschungszentrum Jülich, Germany

Nutrient gradients and limitations play a pivotal role in the life of all microbes, both in their natural habitat as well as in artifical, microfluidic systems. Spatial concentration gradients of nutrients in densely packed cell configurations may locally affect the bacterial growth leading to heterogeneous micropopulations. A detailed understanding and quantitative modeling of cellular behaviour under nutrient limitations is thus highly desirable. We use microfluidic cultivations to investigate growth and microbial behaviour under well-controlled conditions. With a reaction-diffusion type model, parameters are extracted from steady-state experiments with a one-dimensional nutrient gradient. Subsequentially, we employ particle-based simulations with these parameters to predict the dynamical growth of a colony in two dimensions. Comparing the results of those simulations with microfluidic experiments yields excellent agreement. Our modeling approach lays the foundation for a better understanding of dynamic microbial growth processes, both in nature and in applied biotechnology.

BP 18.7 Wed 11:30 H 2013

Sensitivity, dynamics and robustness of extracellular PhrA-signaling in Bacillus subtilis — Heiko Babel[1,2], •Pablo Naranjo[1,2], Stephanie Trauth[1,2], Victor Sourjik[1], and Ilka Bischofs[1,2] — [1]MPI for Terrestrial Microbiology, Marburg, Germany — [2]BioQuant, University of Heidelberg, Germany

Communication is an essential for the self-organization of bacterial populations. The underlying molecular networks that serve this task are surprisingly diverse. In order to understand how the different architectures affect signaling performance, new biophysical tools are required that allow us to quantitatively characterize the function of individual network components and signaling processes in the bacterial cell. A common form of signaling in Gram-positive bacteria is by means of signaling peptides that are produced by an active export-import circuit and are sensed intracellularly. Here, we developed the first FRET-reporter to quantitatively study PhrA-signaling in Bacillus subtilis. Using acceptor photo-bleaching experiments we studied the intra- and extracellular dynamics in response to peptide stimulation and developed a mathematical model that fits the data well. We find that the PhrA signaling circuit, although relying on a low affinity receptor, exhibits exquisite sensitivity to low extracellular signal levels. Our data furthermore suggests that oligopermeases - a component that is shared by all RNPP-signaling circuits in Gram-positive bacteria - play a central role in governing the sensitivity and dynamics of extracellular peptide signaling, while potentially also limiting the robustness of signaling in the presence of other peptides.

BP 18.8 Wed 11:45 H 2013

Modelling of front instabilities in surfactant-driven biofilm spreading — •Sarah Trinschek[1,2], Karin John[2], Sigolène Lecuyer[2], and Uwe Thiele[1,3] — [1]Institut für Theoretische Physik, WWU, Münster, Germany — [2]Université Grenoble-Alpes, CNRS, Laboratoire Interdisciplinaire de Physique, Grenoble, France — [3]Center for Nonlinear Science (CeNoS), WWU, Münster, Germany

The spreading of bacterial colonies at solid air interfaces hinges on physical processes connected to the properties of the involved interfaces. The production of surfactant molecules by the bacteria is one strategy that allows the bacterial colony to efficiently expand over a substrate. These surfactant molecules affect the surface tension which results in an increased wettability as discussed in [1] as well as in outward-pointing Marangoni fluxes that promote spreading. These fluxes may cause an instability of the circular colony shape and the subsequent formation of fingers. In this work, we study the front instability of bacterial colonies at solid-air interfaces induced by surfactant production in the framework of a passive hydrodynamic thin-film model which is extended by bioactive terms. We show that the interplay between wettability and Marangoni fluxes determines the spreading dynamics and decides whether the colony can expand over the substrate. We observe four different types of spreading behaviour, namely, arrested and continuous spreading of circular colonies, slightly modulated front lines and the formation of pronounced fingers.

[1] S. Trinschek et al., PRL 119, 078003 (2017)

BP 18.9 Wed 12:00 H 2013

Pili-mediated substrate motility of bacteria — •Wolfram Pönisch[1], Christoph A. Weber[1,2], and Vasily Zaburdaev[1] — [1]Max Planck Institut für Physik Komplexer Systeme, Dresden, Germany — [2]Paulson School of Engineering and Applied Sciences, Harvard University, Cambridge, USA

Most bacteria live in complex multicellular communities, known as biofilms, colonizing various surfaces. A wide range of bacteria use cell appendages, so called type IV pili, to bind to a substrate and generate pulling forces, enabling the cells to actively move. The attachment of bacteria to a surface and surface associated motility represent the first steps of biofilm formation.

For *Neisseria gonorrhoeae* (NG) bacteria, it was shown that its motility could be described as a persistent random walk with a characteristic length scale that exceeded the average pili length. Previously, it has been suggested that such behavior would require a mechanism of directional memory in pili attachments. Here, we develop a stochastic model demonstrating that the persistent motion arises naturally from the force-dependent detachment rate of pili and the geometric properties of the cell and its pili, but does not require any directional memory of the pili. We confirm this result with the help of a computational model of NG cells interacting with a substrate via its multiple individual pili. Furthermore, in agreement with experimental data, both model describes the dependence of cell motility on the total number of pili per cell.

BP 18.10 Wed 12:15 H 2013

Regulation of cell volume and intracellular biomass density in bacteria, elucidated by single-cell measurements and perturbations — •Enno Oldewurtel and Sven van Teeffelen — Morphogenesis and Microbial Growth Lab, Institut Pasteur, Paris, France

All cells must control their volumes to maintain a high level of macromolecular crowding. In bacteria, cell volume is set by the peptidoglycan (PG) cell wall, which counteracts high internal Turgor pressure. A regulation of PG synthesis and cleavage is required to ensure a match of volume increase with the rate of biomass growth and to prevent cell lysis. We ask: a) How strictly is cell-wall expansion tied to biomass growth? b) Which cell-wall remodeling process is rate-limiting for cell-wall expansion? Using *Escherichia coli* we monitored cell mass and cell dimensions at the single-cell level and over time using quantitative phase microscopy. First, we find control of biomass density both during steady-state growth and during changes in growth rate, suggesting a close coupling of cell-wall expansion with mass growth. Second we show normal volume expansion even if PG synthesis is inhibited, up to the point of cell lysis. Furthermore, transient changes in the rate of PG cleavage lead to rapid changes in the surface expansion rate, pointing to a control of expansion by the rate of PG cleavage. However, despite rapid changes in surface expansion rate, the biomass density stays constant as cell width decreases to maintain a constant volume expansion rate. This width change is likely due to a change in pressure. This suggest cell volume regulation by cell-wall cleavage on intermediate time scales, and an involvement of Turgor pressure on short times.

BP 18.11 Wed 12:30 H 2013

Phenotyping Individual Microbes by Mechanical Stress — •Fabian Czerwinski[1], Bob Fregin[1], Albert Siryaporn[2], and Oliver Otto[1] — [1]Center for Innovation Competence: Humoral Immune Reactions in Cardiovascular Diseases, University of Greifswald, Germany — [2]Department of Physics and Astronomy, University of California Irvine, USA

Microbes typically thrive and prosper in colonies and biofilms, whilst responding very sensitively to their environment. Often, smaller subpopulations take on special tasks that are important for the fate of the bunch. However, probing them individually with high throughput is challenging.

Microfluidics allow for a rapid phenotying of whole bacterial populations ideally capturing individual cells and, therefore, special states. By using the platform of real-time deformability cytometry (RT-DC), e.g., one can distinguish different vegetative states within even huge populations at throughputs beyond 10,000 cells per minute.

We used finite-element simulations to optimize the microfluidic geometries used in RT-DC for geometrical constraints, for flow conditions, and for cellular features. For bacteria, deformation of individual cells as a result of mechanical stress experienced during flow can serve as a distinctive gate.

BP 18.12 Wed 12:45 H 2013

Mechanics of twitching migration of the bacterium P. aeruginosa — •Ahmet Nihat Simsek[1], Matthias D. Koch[2], Benedikt Sabass[1], Gerhard Gompper[1], Zemer Gitai[2], and Joshua W. Shaevitz[2] — [1]Theoretical Soft Matter and Biophysics, Institute of Complex Systems and Institute of Advanced Simulation, Forschungszentrum Juelich, D-52425 Juelich, Germany — [2]Lewis-Sigler Institute for Integrative Genomics, Princeton University, NJ 08544

Pseudomonas aeruginosa is an ubiquitous pathogen responsible for severe and chronic infections. The bacterium employs retractable type-IV pili for migration and colonizes a broad variety of biotic and abiotic surfaces. How surface properties affect migration and colony formation of P. aeruginosa is a potentially important factor for bacterial surface contamination and infections alike. Here, we theoretically and experimentally study the effect of surface properties on the migration of P. aeruginosa. In our model, we assume a rod-like bacterium and treat each pilus, as well as the substrate as elastic springs. Pilus assembly and retraction are modeled as stochastic, force-dependent processes. To generate experimental data, we perform extensive image analysis to record the migration of different strains of P. aeruginosa on polyacrylamide substrates. Experimental data shows a non-linear dependence of migration speed and mean square displacement on substrate properties, which is in accordance with simulations. Finally, we present an analytical theory including the calculation of effective diffusion coefficients and aggregation probabilities of bacteria.