Biology Across Scales

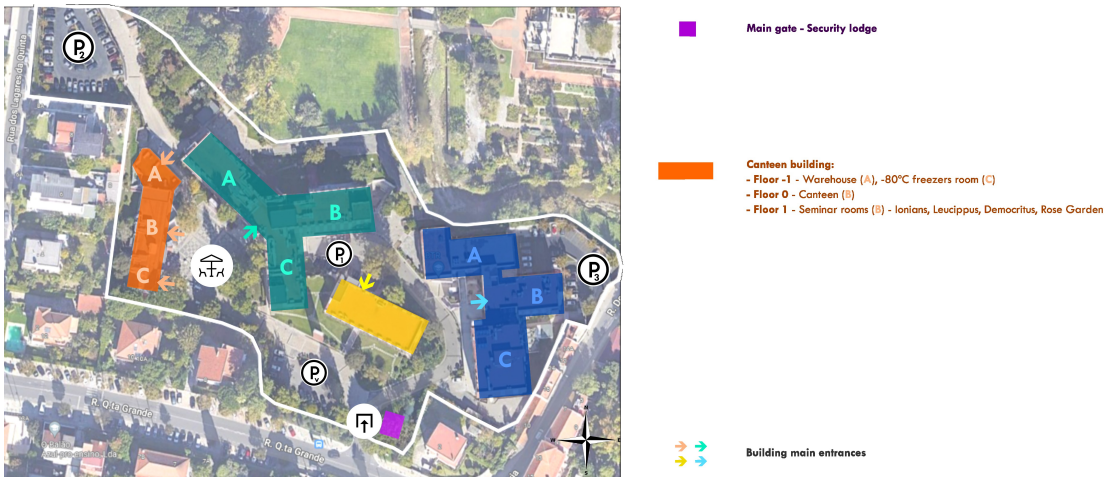
Book of abstracts

GULBENKIAN
CIÊNCIA

Useful information

Conference venue

The Biology Across Scales conference will take place at the Instituto Gulbenkian de Ciência (IGC). Here we provide a map of the IGC campus:



The conference will take place in the Canteen building (indicated in orange in the map). Scientific activities will take place in the first floor of the building: talks will take place at the Ionians auditorium, while poster sessions will take place at the Leucippus and Democritus seminar rooms. Lunch will be provided at the IGC Canteen in the ground floor.

Internet access

Wifi access at the IGC is available through the eduroam or IGC networks. To access the IGC network provide the password chemokine.

Conference dinner

The conference dinner will take place at 20:00 on Thursday the 21st of March at the Azure restaurant, approximately 25 minutes away by foot from the IGC campus.

Programme

Wednesday - 20/03/2024

09:00 - 09:20 Opening remarks

Session I

09:20 - 09:50 Giulia Ghedini (IGC) - Metabolic responses to competition explain the origin of community scaling patterns
09:50 - 10:20 Jacopo Grilli (ICTP) - Dynamics of microbial communities, in-vivo and in-silico
10:20 - 10:40 Lilla Lovasz (U. Basel) - Effects of large herbivores on the functional composition of vegetation in a restored wetland ecosystem
10:40 - 11:00 Onofrio Mazzarisi (ICTP) - Diversity begets stability: sublinear growth and competitive coexistence across ecosystems

11:00 - 11:30 Coffee break

Session II

11:30 - 12:00 Claudia Bank (U. Bern) - Fitness landscapes for the study of ecology and evolution across biological scales
12:00 - 12:20 Paula García-Galindo (U. Cambridge) - The non-deterministic genotype–phenotype map of RNA secondary structure
12:20 - 12:40 Saúl Huitzl (Northwestern U.) - Evolutionary Origins of Hierarchical Organization in Biological Systems

12:40 - 14:00 Lunch break

Keynote speaker

14:00 - 15:00 Ian Hatton (McGill U.) - Similar structural and dynamical scaling from ribosomes to whole biomes

Session III

15:00 - 15:30 Guillaume Achaz (U. Paris Cité) - Weak genetic draft and the Lewontin's paradox

15:30 - 15:50 David Soriano-Paños (U. Rovira i Virgili) - Impact of life-history traits on the genetic fingerprint of habitat changes
15:50 - 16:10 Dennis Lavrov (Iowa State U.) - Exploring the connection between unusual mt-genome organization, peculiar population biology of calcaronean sponges, and mRNA editing in their mitochondria

16:10 - 18:00 Poster session (focus on odd numbers)

Thursday - 21/03/2024

Session IV

09:00 - 09:30 Michi Taga (U. California Berkeley) - The corrinoid model illuminates cross-scale microbial community interactions
09:30 - 10:00 Massimo Amicone (U. Lausanne) - Leveraging ecological principles to control microbial ecosystems: assembly, function and evolution
10:00 - 10:20 Audrey Menegaz Proença (Freie U. Berlin) - A tiny form of aging that can determine life and death
10:20 - 10:40 Mónica Louro (IGC) - Biofilm formation in multicellular gut microbiota communities
10:40 - 11:00 José Manuel Camacho Mateu (UC3M) - Sparse species interactions reproduce abundance correlation patterns in microbial communities

11:00 - 11:30 Coffee break

Session V

11:30 - 12:00 Lucie Etienne (ENS Lyon) - Genomic and functional diversification of the bat innate immunity in response to viral infections
12:00 - 12:20 Chujin Ruan (Eawag/ETH Zurich) - Phage predation accelerates the spread of plasmid-encoded antibiotic resistance
12:20 - 12:40 Nelson Frazão (IGC) - The impact of pregnancy on gut microbiota evolution

12:40 - 14:00 Lunch break

Keynote speaker

14:00 - 15:00 Sylvia Cremer (ISTA) - Social immunity: disease protection in social insect colonies

Session VI

15:00 - 15:30 Simon Babayan (U. Glasgow) - Environmental drivers of low vaccine responsiveness in a lab-to-wild rodent model
15:30 - 15:50 Alexandra Kortsinoglou (U. Athens) - Investigating Endophytism in Entomopathogenic Fungi: A Comprehensive Exploration through Comparative Genomics
15:50 - 16:10 Abhishek Nair (U. Ahmedabad) - Altitudinal variations in cuticular hydrocarbons in Drosophila melanogaster from the Western Himalayas

16:10 - 18:00 Poster session (focus on even numbers)

20:00 - 23:00 Conference dinner at Azure

Friday - 22/03/2024

Session VII

09:00 - 09:30 Saúl Ares (CNB-CSIC) - Feedback control of organ size precision is mediated by apoptosis in the Drosophila eye
09:30 - 10:00 Roberto Arbore (CIBIO) - The bio-architecture of feather colours
10:00 - 10:20 Natalia Lavalle (U. La Plata/CONICET) - Local control of cellular proliferation during organ regeneration in zebrafish
10:20 - 10:40 Ali Seleit (EMBL) - Modular control of time and space during vertebrate axis segmentation
10:00 - 10:20 Bruno Martins (U. Warwick) - The cyanobacterial circadian clock couples to pulsatile processes using pulse amplitude modulation

11:00 - 11:30 Coffee break

Session VIII

11:30 - 12:00 Marco Cosentino Lagomarsino (IFOM/U. Milan) - Are there "universal" laws of cellular growth?
12:00 - 12:20 Damiano Andreghetti (Polytechnic U. Torino) - Molecular sorting on a fluctuating membrane
12:20 - 12:40 Victor Hugo Mello (IGC) - From structural variability to protein energetics of a molecular motor

12:40 - 14:00 Lunch break

Keynote speaker

14:00 - 15:00 Denis Duboule (Collège de France/EPFL) - Using pseudo-embryos to study the Hox timer

Session IX

15:00 - 15:30 Dimitrios Stavropodis (U. Athens) - 1993 - 2023: 30 Years of Cytokine Signaling
15:30 - 15:50 Zahra Eidi (IPM) - Correspondence between multiple signaling and developmental cellular patterns: a computational perspective
15:50 - 16:10 Kesavan Subburam (Lund U.) - Observing DNA damage responses of cells with Data-Driven Microscopy

16:10 - 16:40 Coffee break

Session X

16:40 - 17:00 Carolina Pereira (IGC) - Understanding mitotic physiology of pluripotent stem cells
17:00 - 17:20 Nuno Santos (IMM) - Interactions across scales: protein-erythrocyte

and erythrocyte-erythrocyte binding as determinants for cardiovascular risk
17:20 - 17:40 Closing remarks

18:00 - 22:00 Beer hour at the IGC patio

Abstracts - Speakers

Metabolic responses to competition explain the origin of community scaling patterns

20 Mar
09:20-09:50

Giulia Ghedini
IGC

Metabolism and growth scale sublinearly with body mass for most species across the tree of life. Ecosystems show a similar sublinear scaling between biomass production and total biomass. But ecological theory cannot reconcile the existence of nearly identical scalings at different levels of biological organization. I will present recent work that solves this paradox by challenging a key assumption of current theory: that metabolic allometries measured for species in isolation reflect those of species in communities. Using marine phytoplankton as a model system, we discovered that competition slows metabolism and production at a unique rate across species shifting the scaling of metabolism with size in a predictable fashion. By accounting for this effect, we reconcile individual and community scaling patterns. To prove the generality of this new principle, we worked across communities spanning three orders of magnitude in species size and biomass, five resource regimes, and systems both far and close to equilibrium. We demonstrate that the same metabolic regulation operates across species and systems. These results identify the origin of ecosystem allometries and unite aspects of physiology and ecology to explain why growth patterns are so strikingly similar across scales.

Dynamics of microbial communities, in-vivo and in-silico

20 Mar
09:50-10:20

Jacopo Grilli
ICTP

Thousands of microbial species coexist in the human gut or in a gram of soil, but what maintains community diversity is still unclear. Experiments show that tens of species can coexist under simple conditions, e.g., where a single carbon source is provided. The complexity of the structure of these communities reflects the hidden complexity of the network of inter-species interactions and of the chemical and physical environment, which are constantly altered by bacterial growth phases. In this talk, I will discuss how the statistical properties of environmental variability can give rise to regularities in community variation. By studying this connection, I will show how relatively simple models of stochastic population dynamics can capture patterns of variability in empirical and experimental communities.

Effects of large herbivores on the functional composition of vegetation in a restored wetland ecosystem

20 Mar
10:20-10:40

Lovasz, Lilla (1,2), Korner-Nievergelt, Fränzi (3), Amrhein, Valentin (1,2)

(1) Zoological Institute, University of Basel, Switzerland; (2) Research Station Petite Camargue Alsacienne, Saint-Louis, France; (3) Oikostat GmBH, Ettiswil, Switzerland

Rewilding initiatives in European open and semi-open lowlands increasingly involve domestic cattle and horses for ecological restoration, especially in wetland areas of high conservation value. These large herbivores contribute to spatial heterogeneity and enhance biodiversity by shaping ecosystems through movement, grazing, and resting behaviours. However, their site-specific habitat use patterns and impact on plant communities remain unclear. In this study, we investigated the spatiotemporal distribution of free-roaming cattle and horses in a French nature reserve, assessing varying habitat use intensity. We explored differences in habitat use between the two species during summer and winter on a macrohabitat scale. Additionally, we examined structural and functional changes in vegetation traits over four years, focusing on plant height, patch cover, species abundance, and functional traits. The study site, a former agricultural area converted into an ecological restoration site, allowed observation of ecological processes from a "zero state." Findings suggest that cattle and horses exhibit similar habitat choices with seasonal variations, potentially indicating shared feeding niches. Despite concerns about potential negative effects, multi-species grazing had a sustaining effect on plant cover and vegetation height without causing destructive impacts. The herbivores' presence led to slight changes in abundance of light-preferent and nutrient-tolerant species and an increase in grazing-tolerant plants. This study contributes insights into the seasonal and interspecific variability of habitat selection by large herbivores and vegetation dynamics in a rewilding-based ecosystem restoration project, informing management strategies for conservation initiatives.

Diversity begets stability: sublinear growth and competitive coexistence across ecosystems

20 Mar
10:40-11:00

Ian A. Hatton, Ian A. (1, 2); Mazzarisi, Onofrio (1, 3); Altieri, Ada (4); Smerlak, Matteo (1, 5, 6)

(1) Max Planck Institute for Mathematics in the Sciences, Leipzig, Germany; (2) Department of Earth and Planetary Sciences, McGill University, Montreal, Canada; (3) The Abdus Salam International Centre for Theoretical Physics (ICTP), Trieste, Italy; (4) Laboratoire Matière et Systèmes Complexes (MSC), Université Paris Cité CNRS, France; (5) Laboratoire de Biophysique et Evolution, UMR 8231 CBI, ESPCI Paris, PSL Research University, France; (6) Capital Fund Management, Paris, France

The dramatic loss of species diversity brings urgency to understanding how diverse ecosystems maintain stability. Whereas early ecological ideas and classic observations suggest that stability increases with diversity, ecological theory makes the opposite prediction, leading to the longstanding "diversity-stability debate". Here we show this puzzle is resolved if growth scales as a sublinear power law with biomass

(exponent < 1), exhibiting a form of population self-regulation analogous to models of individual ontogeny. We show that competitive interactions among populations with sublinear growth do not lead to exclusion, as occurs with logistic growth, but instead promote stability at higher diversity. Our model realigns theory with classic observations and predicts large-scale macroecological patterns. However, it makes an unsettling prediction: biodiversity loss may accelerate the destabilization of ecosystems.

Fitness landscapes for the study of ecology and evolution across biological scales

20 Mar
11:30-12:00

Claudia Bank

U. Bern

Fitness landscapes are maps that describe the relationship between genotypes, phenotypes, and/or fitness. Initially proposed as a theoretical concept almost a century ago, the theory of fitness landscape has been applied at multiple biological scales in the context of research in adaptation, speciation, molecular evolution, systems biology, etc. Moreover, in the past two decades, increasingly large experimental fitness landscapes have been screened, primarily at the within- or between-gene level. I will highlight examples of published and ongoing fitness landscape research across biological scales. I will use these examples to discuss to which extent the concept indeed captures general patterns across biological levels and what fitness landscape research teaches us about evolution.

The non-deterministic genotype–phenotype map of RNA secondary structure

20 Mar
12:00-12:20

García-Galindo, Paula (1) Ahnert, Sebastian E. (1,2) Martin, Nora S. (3)

(1) Department of Chemical Engineering and Biotechnology, University of Cambridge, UK (2) The Alan Turing Institute, London, UK (3) Rudolf Peierls Centre for Theoretical Physics, Oxford, UK

Selection and variation are both key aspects in the evolutionary process. Previous research on the mapping between molecular sequence (genotype) and molecular fold (phenotype) has shown the presence of several structural properties in different biological contexts, implying that these might be universal in evolutionary spaces. The deterministic genotype–phenotype (GP) map that links short RNA sequences to minimum free energy secondary structures has been studied extensively because of its computational tractability and biologically realistic nature. However, this mapping ignores the phenotypic plasticity of RNA. We define a GP map that incorporates non-deterministic (ND) phenotypes, and take RNA as a case study; we use the Boltzmann probability distribution of folded structures and examine the structural properties of ND GP maps for RNA sequences of length 12 and coarse-grained RNA structures of length 30 (RNAshapes30). A framework is presented to

study robustness, evolvability and neutral spaces in the ND map. This framework is validated by demonstrating close correspondence between the ND quantities and sample averages of their deterministic counterparts. When using the ND framework we observe the same structural properties as in the deterministic GP map, such as bias, negative correlation between genotypic robustness and evolvability, and positive correlation between phenotypic robustness and evolvability.

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20 Mar
12:20-12:40

Evolutionary Origins of Hierarchical Organization in Biological Systems

Saúl Huitzil (1,2), Cristián Huepe (1,2,3)

(1) Engineering Sciences and Applied Mathematics, Northwestern University, Evanston, USA, (2) Northwestern Institute on Complex Systems, Northwestern University, USA, (3) CHuepe Labs, Chicago, USA

Biological systems exhibit hierarchical organization across multiple scales, from proteins to ecologies. Additionally, each system is composed of interconnected subsystems, developing structures at all levels. Our understanding of how evolution led to this form of organization and its implications remains limited. One hypothesis is that modularity allows for the recombination of simpler subparts into a broad library of increasingly complex systems. Cells form different types of tissues, which recombine to create different organs, then organisms, etc. up to the ecological scale. In this study, we introduce an evolutionary Boolean network model where nodes represent the components of a given evolving system, and connections are the interactions between them. The state of each node is computed as the output of its Boolean function, with inputs given by its incoming links. We define a subset of nodes as the inputs of the network system, and one node, as its output. We evolve the links and Boolean functions of a population of networks, aiming for the network output to match a predefined fitness function. Adaptation thus occurs at two levels: at the component (node) level, through its Boolean functions, and at the system level, through the network links. Our results demonstrate that modularity and hierarchical organization can optimize different forms of adaptation, spontaneously emerging for a broad range of conditions. Structural and functional modules can recombine at multiple levels to tackle more tasks of increasing complexity, leading to the multiscale hierarchical modularity observed in living systems.

Similar structural and dynamical scaling from ribosomes to whole biomes

20 Mar
14:00-15:00

Ian Hatton
McGill University

There are two opposing explanatory paradigms in biology. Below the level of the individual, cells, tissues and organs are thought to serve the higher level functioning of the individual. Above the level of the individual, on the other hand, populations, communities and ecosystems are thought to emerge from individual behaviours. While both explanatory paradigms originate with the individual, these two forms of causation point in opposite directions (top-down teleology vs. bottom-up emergence). Here we show three different classes of scaling patterns that encompass biological structure, variation and dynamics, and span an enormous range, from ribosomes to whole biomes. These scaling patterns show remarkable similarity both below and above the level of the individual. While interesting in their own right, their striking parallels across levels of organization raises a problem: the same pattern requires two opposing explanations. We discuss the implications of these patterns for biological theory.

Weak genetic draft and the Lewontin's paradox

20 Mar
15:00-15:30

Guillaume Achaz
U. Paris Cité

Neutral theory assumes that in a population of size N, diversity results from an equilibrium between new mutations arising at rate μ and genetic drift that purge them at rate $1/N$, predicting an equilibrium value proportional to $N\mu$. The difference between this expectation and the much lower observed molecular diversity is known as the Lewontin's paradox of variation. Here, we investigate the effect of genetic draft, a regime of evolution where recurrent sparse selective sweeps entirely drive the diversity of surrounding loci. More specifically, we focus on the neglected distant effect of selective sweeps on remote neutral loci, where the effect of a single sweep is almost negligible. We derived novel mathematical approximations of this underexplored regime and show that under weak genetic draft, diversity at neutral loci is a power law of the population size: $N_e = cstN^{2A}$, for $A < 0.5$, where A is the ratio between recombination rate and coefficient of selection ($A = c/s$). Interestingly the Site Frequency Spectrum at neutral loci is identical to the one produced by genetic drift, as the underlying coalescent tree is an n-Kingman coalescent. In brief, weak genetic draft produces patterns of diversity that look entirely neutral, while being drastically reduced in magnitude. Ultimately, our study points to the need to explore evolutionary models for which diversity looks neutral but does not scale linearly with population size.

Impact of life-history traits on the genetic fingerprint of habitat changes

20 Mar
15:30-15:50

Vishwakarma, Ravi (1); Sgarlata, Gabriele (1); Soriano-Paños, David (1,2); Rasteiro, Rita (1,3); Maié, Tiago (1,4); Paixão, Tiago (1); Tournebize, Rémi (5); Chikhi, Lounes (1,5,6)

(1) Instituto Gulbenkian de Ciência, Portugal; (2) Institute for Biocomputation and Physics of Complex Systems, University of Zaragoza, Spain; (3) School of Biological Sciences, University of Bristol, Bristol, United Kingdom; (4) Institute for Computation Genomics, RWTH Aachen University, Germany; (5) Laboratoire Évolution Diversité Biologique, CNRS, IRD, UPS, Université de Toulouse Midi-Pyrénées, France; (6) Centre de Recherche sur la Biodiversité et l'Environnement, CNRS, IRD, UPS, Université de Toulouse Midi-Pyrénées, France.

Species ranges are dynamic, experiencing expansions, contractions or shifts as a response to habitat changes induced by extrinsic factors such as climate change and, more recently, human activities. While the scientific literature has explored the genetic effects of spatial processes, published studies rarely incorporate life-history traits to study the effect of such changes on species living in the same environments. There is thus a gap in our understanding regarding the variation in genetic diversity patterns among species with distinct life-history traits such as growth rates and generation times, experiencing the same habitat change scenarios. In this study, we first used spatial simulations to investigate the temporal dynamics of genetic diversity within refugium populations experiencing a range expansion followed by a stationary and a contraction period. We explored different scenarios, varying both the speed of contraction and the life-history traits of the simulated species. In addition we used a simpler panmictic model for which we derived analytical results. Altogether, we identified three temporal dynamics of genetic diversity in the refugium population during the contraction phase: scenarios where genetic diversity (i) decreased throughout the contractions, (ii) increased for periods that could be greater than thousands of years before plateauing and then decreasing or (iii) followed a persistent increasing trend, without any visible effect of the expansion or contraction despite the fact that census size increased and then decreased by two orders of magnitude in all scenarios.

Exploring the connection between unusual mt-genome organization, peculiar population biology of calcaronean sponges, and mRNA editing in their mitochondria

20 Mar
15:50-16:10

Lavrov, Dennis V. (1)

(1) Dept. of Ecology, Evolution, and Organismal Biology, Iowa State University

Transcripts of mitochondrial genes in calcaronean sponges undergo an unusual process of insertional mRNA editing in which 1-3 uridylates are inserted at specific sequence motifs. Each coding sequences can contain up to 36 edited sites and failure of editing of any one of them would lead to a frameshift and a non-functional

protein. Despite such strong functional constraints, editing sites are not well conserved in evolution and the rate of their appearance/disappearance is comparable to those of single nucleotide substitutions. Here I argue that evolution of editing sites in these sponges is enabled by highly unusual mitogenome organization in these species, where each gene is located on a separate linear chromosome and multiple copies of each chromosome is transmitted from generation to generation. Furthermore, I suggest that this unusual genome organization is an inadvertent result of the unusual population and reproductive biology of most species in the group, with many populations experiencing annual boom and bust cycles. Overall, this group provides a stark example on how ecology and life histories of a species can influence its genome organization and molecular evolution.

The corrinoid model illuminates cross-scale microbial community interactions

21 Mar
09:00-09:30

Michi Taga

U. California Berkeley

Nutritional interactions are widespread among microbes and are major drivers of microbial community structure. The long-term goal of my research program is to understand microbial interactions across scales, from molecular mechanisms of interactions to their impact on communities. We have developed the corrinoid model to study nutrient-sharing interactions among microbes across scales. Corrinoids are the vitamin B12 (cobalamin) family of cofactors, involved in diverse metabolic processes in prokaryotes and essential for humans. Importantly, corrinoids are produced by only a fraction of prokaryotes, suggesting they are widely shared metabolites. Further, corrinoids are structurally diverse, and corrinoid-dependent organisms have specificity for particular corrinoids. At the molecular level, we found that corrinoid specificity in bacterial growth can be attributed to selectivity in corrinoid-dependent enzymes and corrinoid-responsive regulatory RNA elements (riboswitches). At the community level, addition of certain corrinoids can transiently shift bacterial composition, even in the complex microbiome of soil. We are using a combination of bioinformatic, genetic, and microbiological approaches to discover how the interactions between corrinoid-producing and -requiring microbes evolve and shape community structure.

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21 Mar
09:30-10:00

Leveraging ecological principles to control microbial ecosystems: assembly, function and evolution

Massimo Amicone

U. of Lausanne

Microbes influence the health of every ecosystem on Earth. Achieving full control over microbial communities has limitless potential, from habitat conservation to bioremediation and therapeutics. Yet, our ability to control and/or reproduce microbial ecosystems is limited by the lack of fundamental understanding of how they assemble, function, and evolve. Combining mathematical modeling with experiments, research in the Mitri lab aims to leverage ecological principles to achieve full control over microbial communities, from synthetic ecosystems to more natural ones. Among the most complex and often neglected features of microbial communities is their evolution. Overall, we now understand relatively well how different species can form a functional community, less how these evolve. My research focuses on the interplay between ecological and evolutionary processes shaping species-rich communities. Here, I will present an ongoing project whereby we look at the effects of ecological interactions on the emergence of resistance to antibiotics or other toxins. By focusing on a two-species ecosystem, we identify a key factor determining the likelihood of the susceptible species to acquire resistance: the balance between competition and facilitation (via environmental detoxification) with the partner species. We test predictions stemming from mathematical models through in vitro experimental evolution of bacteria and show how merging the ecological and evolutionary scales is key to control the response of microbial ecosystems to environmental challenges.

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21 Mar
10:00-12:20

A tiny form of aging that can determine life and death

Proença, Audrey M. (1); Tugrul, Murat (1); Nath, Arpita (1); Steiner, Ulrich (1)

(1) Evolutionary Demography Group, Institute of Biology, Freie Universität Berlin, Germany

Aging is a progressive functional decline over the lifetime of an organism. In complex organisms, this is often represented by a parent that accumulates damage over time, retaining this damage to produce rejuvenated offspring. Nonetheless, recent microscopy advances showed that unicellular organisms such as bacteria and yeast, and algae are not exempt from aging. When rod-shaped bacteria reproduce, a mother cell inherits an old pole carrying damage accumulated across generations, thus aging. The daughter cell it produces is born rejuvenated, because it inherits newly synthesized poles carrying less damage. What are the consequences of this simple form of aging for the individual and the population? We demonstrated that Escherichia coli cells age and rejuvenate even when grown under stable environmental conditions, showing that aging represents a deterministic source of variability for bacteria. This asymmetry reaches a stable equilibrium despite the large stochasticity natural to bacterial growth and division. Although damage inheritance plays

a large role in defining aging and rejuvenation, we found that asymmetry is also present in single-cell gene expression. Mother cells show lower rates of gene expression than their daughters, developing an intracellular expression gradient as the old pole ages over generations. Finally, we showed that aging and rejuvenation can be a matter of life and death when facing environmental stresses: under oxidative stress, mother cells cross a mortality threshold while their daughters can continue to proliferate. Thus, aging in its simplest form acts as a driver of phenotypic heterogeneity in bacterial populations, with consequences for stress survival.

Biofilm formation in multicellular gut microbiota communities

21 Mar
10:20-10:40

Louro, Mónica (1); B. Xavier, Karina (1)

(1) Instituto Gulbenkian de Ciência

Bacteria can produce extracellular polymers to form a matrix to which cells aggregate, forming multicellular structures called biofilms. Biofilms have been studied on single-species pathogenic bacteria, such as Pseudomonas aeruginosa and Staphylococcus aureus and are often associated with disease. These structures can confer multiple advantages to bacteria, such as prolonged permanence inside a host or abiotic surface, close association with other bacterial cells for exchange of nutrients and DNA, higher tolerance to external insults like antibiotics, and host responses. Recently, it has been proposed that biofilm formation by gut symbionts could be an important mechanism that increases resilience of these bacteria to perturbations that can affect colonization of gut microbiota symbionts, such as diet and antibiotics. However, the formation of biofilms in gut symbionts has been studied only using mono-species cultures. As the mammalian gut is composed of hundreds of different species interacting with each other, we propose to study biofilms in multi-species cultures. To study the role of biofilm formation in gut symbionts our lab is using a defined community, the Oligo-Mouse Microbiota 12 (OMM12). Our results indicate that Enterococcus faecalis, a member of OMM12, has a key role in the formation of community biofilms, namely as the major matrix producer. Future work will investigate the molecular mechanisms involved in the key role of E. faecalis in the formation of OMM12 multi-species biofilms. Moreover, we will determine the relevance of these mechanisms in how gut microbiota communities respond to environmental perturbations.

Sparse species interactions reproduce abundance correlation patterns in microbial communities

21 Mar
10:40-11:00

(1) Jose Camacho-Mateu, (1) Aniello Lampo, (2) Matteo Sireci, (2) Miguel A Muñoz, (2) Jose A Cuesta

(1) Carlos III University of Madrid ; (2) University of Granada

Microbes—the most abundant organisms on Earth—form complex communities with a profound influence on many problems, from human health to ecosystem management. Understanding species interactions is of paramount importance in many different contexts, e.g., in medical applications. However, current models of microbial communities have not adequately captured the essential role of interactions. Here, we present population models implementing species interactions. These enable us to replicate macroecological patterns of species correlations not captured by existing models. These findings robustly support the importance of species interaction networks, highlighting their inherent sparsity as a distinctive structural property. This characteristic is associated with the prevalence of amensalistic and commensalistic relationships within the community.

Genomic and functional diversification of the bat innate immunity in response to viral infections

21 Mar
11:30-12:00

Lucie Etienne

ENS Lyon

Bats (Chiroptera) are a diverse and widespread mammalian order. They harbor a wide variety of viruses, including zoonoses, and appear asymptomatic to infections that are pathogenic to most mammals. Their life-history traits, physiology and immunity seem to participate to these original features. Comparative genomic, transcriptomic, and functional studies have inferred that bats have evolved a balance between viral tolerance and antiviral defense. Yet, most of the functional evolutionary history of bat antiviral immunity is unknown. Because innate immunity is the first line of antiviral defense and the link with inflammation and adaptive immunity, we aimed at characterizing its response. We first used genome-wide and gene candidate evolutionary analyses and found that many antiviral effectors have undergone lineage-specific adaptation and duplications, notably in Myotis bats. Thanks to an international research collaboration, we combined ecology, novel genomic sequences and primary cells from 18 individuals from six Myotis species, with viral infections, omics, evolutionary and mechanistic studies. We notably found that infections of Myotis primary cells with diverse viruses showed species- and virus-specificity. Upon immune stimulation, most cells were capable of mounting an antiviral response, albeit with strong differences in the magnitude of restriction. Through transcriptomics, we identified the core innate immune response, as well as the specific quantitative and qualitive differences. We are now characterizing some adaptive duplications and innovations in bat immunity, providing insights on its functional evolution and modern consequences. Identifying key virus-host interfaces

may further inform prevention and therapeutic strategies and unveil factors involved in cross-species transmissions.

Phage predation accelerates the spread of plasmid-encoded antibiotic resistance

21 Mar
12:00-12:20

Ruan, Chujin(1,2); Ramoneda, Josep(3); Kan, Anton(4); Rudge, Timothy J(5); Wang, Gang(2,6); Johnson, David R(1,7).

(1)Department of Environmental Microbiology, Swiss Federal Institute of Aquatic Science and Technology (Eawag), Switzerland; (2)College of Land Science and Technology, China Agricultural University, Beijing, China; (3)Cooperative Institute for Research in Environmental Sciences, University of Colorado, USA; (4)Department of Materials, Swiss Federal Institute of Technology (ETH), Zürich, Switzerland; (5)Interdisciplinary Computing and Complex Biosystems (ICOS) Research Group, School of Computing, Newcastle University, UK; (6)National Black Soil and Agriculture Research, China Agricultural University, Beijing, China; (7)Institute of Ecology and Evolution, University of Bern, 3012 Bern, Switzerland

Phage play a central role in directing the structure, functioning and evolutionary trajectories of microbial ecosystems, with significant implications across diverse sectors including human health, biotechnology, biogeochemical cycling and environmental remediation. In the field of phage therapy, phage are used as biological agents to target bacterial populations that have developed resistance to conventional antibiotics. It is generally assumed that the use of predatory phage does not contribute to the spread of antibiotic resistance. Here, we challenge this assumption by providing evidence that phage predation can create conditions conducive to the spread of antibiotic resistance, particularly in scenarios involving surface-associated microbial growth. We conducted experiments using two strains of Escherichia coli where one is a donor of plasmid-encoded antibiotic resistance and the other a potential recipient. We showed that phage predation impedes the spatial segregation of the bacterial strains, which consequently increases intermixing and the frequency of direct cell-to-cell interactions, thereby amplifying the rate and extent of plasmid-mediated transfer of antibiotic resistance. The underlying mechanism is that phage predation shifts the location of maximal bacterial growth from the outer edges of the bacterial biomass towards its interior. This relocation to a more confined space restricts bacterial movement, resulting in the formation of straighter interfaces between the bacterial strains. These straighter interfaces are less prone to merge with adjacent interfaces, which consequently slows the process of spatial segregation and increases the probability of plasmid transfer. In conclusion, our study reveals two important consequences of phage predation: i) it can promote the spread of antibiotic resistance with potential implications for phage therapy, and ii) it provides a mechanism for how costly functions that are deleterious for human and environmental health can proliferate in the environment.

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21 Mar
12:20-12:40

The impact of pregnancy on gut microbiota evolution

Bom, J (1); Seixas, E (1); Frazão, N (1)
(1) Instituto Gulbenkian de Ciência

The distinct physiological conditions imposed by pregnancy are known to induce ecological changes among gut bacteria. However, the evolution of bacteria under pregnancy remains unknown. This study aims at elucidating the genetic evolution of Escherichia coli (E. coli), a canonical bacterium of the mammalian gut microbiota, during pregnancy. Employing a gut colonization approach, we isolated E. coli populations from the gut of both pregnant and non-pregnant germ-free mice, subsequently subjecting them to whole-genome sequencing. Our analysis revealed that a unique adaptive mutation emerged exclusively within E. coli populations isolated from pregnant mice. This intergenic mutation is associated with the import of cysteine, a crucial amino acid with bacterial cytotoxic properties. We hypothesize that it confers a selective advantage by lowering cysteine uptake, safeguarding E. coli from cysteine-induced toxicity during pregnancy and subsequent transmission to the newborn's gut. Importantly, reduced cysteine levels in the bacterial cell can increase the formation of membrane vesicles (MVs) released from the outer membrane of bacteria. MVs can cross the intestinal epithelial barrier, enter the bloodstream, and subsequently access the embryo during pregnancy. Indeed, using electron microscopy we observed MVs within the mouse embryo environment, which may impact development of the immune system. Our study provides new insight into the adaptive evolutionary mechanism of E. coli during pregnancy and its potential impact on the mammalian embryo's immune system before birth.

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21 Mar
14:00-15:00

Social immunity: disease protection in social insect colonies

Sylvia Cremer
ISTA

Social insects like ants, bees and termites are protected against disease not only by the individual immune systems of colony members, but also by their cooperative behaviors that reduce the risk of infection and disease transmission and thereby provide "social immunity". Whole-colony protection is thus achieved by a tight interaction of individual and collective actions of mutual sanitary care and cooperative infection treatment. Little is known, however, about the individual decision rules that trigger these behaviors in colony members and how they interplay to an effective disease control at the colony level. It is also still underexplored how the drastic reduction of pathogen fitness by social immunity affects pathogen adaptation to withstand the colony-level defenses of their social hosts. I present the integrated approach we use to study how social immunity emerges from individual actions and social interactions and how it may shape disease evolution.

Environmental drivers of low vaccine responsiveness in a lab-to-wild rodent model

21 Mar
15:00-15:30

Simon A. Babayan (1), Saudamini Venkatesan (2), Jessica Hall (2), Ewan Smith (1), Amy Sweeny (2), and Amy B Pedersen (2)

(1) School of Biodiversity, One Health and Veterinary Medicine, University of Glasgow, Glasgow, UK (2) Institute of Evolutionary Biology, University of Edinburgh, Edinburgh UK

Vaccination is the most effective way to prevent infectious disease and protect public health. However, most new vaccines fail late into the clinical trial pipeline, and even established vaccines often perform worse in populations that differ substantially from those in which they were initially tested, such as rural vs. urban individuals. This can lead to breakthrough infections and the inability to achieve or maintain herd immunity. We therefore hypothesised that the leading causes of vaccine hyporesponsiveness, are environmental. Using paired cohorts of laboratory-reared and wild wood mice (Apodemus sylvaticus), we show that circulating vaccine-specific antibodies dropped on average by 50% in a wild population compared to individuals of the same species reared in laboratory conditions. We then used structural causal models to disentangle the drivers of the loss of efficacy. We found that, independently of habitat, substantial variation in vaccine responsiveness was explained by diet, with weaker negative impacts of being male and reproductively active. Surprisingly, helminth infection did not directly suppress vaccine-specific antibody production once adjusted for habitat, diet, and sex. While much variation remains to be explained, our results indicate that the environment plays a dramatic role in vaccine failure and that laboratory settings may systematically overestimate vaccine performance by failing to capture natural environmental variability.

Investigating Endophytism in Entomopathogenic Fungi: A Comprehensive Exploration through Comparative Genomics

21 Mar
15:30-15:50

Kortsinoglou, Alexandra (1); Myridakis, Antonis (1); Wood, Martyn (2,3); Butt, Tariq (2); Kouvelis, Vasilli (1)

(1) National And Kapodistrian University Of Athens, Department Of Biology, Section Of Genetics And Biotechnology, Athens, Greece; (2) Swansea University, Swansea, Wales, UK; (3) Institute of Molecular Biology and Biotechnology, Foundation for Research and Technology, Heraclion, Crete, Greece

Entomopathogenic fungi (EPF) are widely acknowledged for their effectiveness as biological control agents against various pests. Additionally, these fungi have a more complex mode of life since they influence plant growth and provide plant protection as endophytes and rhizosphere colonizers. Their dual role makes them promising candidates for developing innovative biocontrol products and biofertilizers. However, limited understanding of the mechanisms behind these abilities and the evolutionary pathways implicated in these interactions impedes progress in this field. This study emphasizes the importance of genomics in unraveling the intricate mechanisms of endophytism in entomopathogenic fungi. Using in-house Oxford Nanopore sequencing,

we conducted whole genome sequencing of Metarhizium brunneum strains ARSEF 4556 and V275, Beauveria bassiana ATHUM 4946, Metarhizium guizhouense ARSEF 819, and Beauveria brongniartii ARSEF 9452. The resulting high-quality whole genomes underwent functional annotation, confirming the versatile gene content and structure of entomopathogenic fungal genomes. Notably, our findings revealed a broad secondary metabolic repertoire in all strains, enabling them to adopt diverse modes of life. Comparative genome analyses unveiled shared orthologue genes with other endophytic and entomopathogenic strains of the Hypocreales order, along with several singleton genes. Phylogenetic trees based on matrices of these genes, provide valuable knowledge into the evolutionary relationships among these fungi. In summary, our comparative genome analyses offer valuable insights into the genomic diversity and evolutionary history of these Hypocrealean EPF strains. Overall, our findings illuminate the evolution of mechanisms and metabolic pathways supporting the endophytic lifestyle in EPF, providing answers to questions about the multifaceted nature of these species' life strategies.

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21 Mar
15:50-16:10

Altitudinal variations in cuticular hydrocarbons in Drosophila melanogaster from the Western Himalayas

(1) Nair, Abhishek (2) Mayekar, Harshad (3) Sharma, Manmohan (4) Garg, Divita (5) Mitchell, Christopher (5) Hosken, David J (6) Rajpurohit, Subhash

(1) Division of Biological Sciences, School of Arts and Sciences, Ahmedabad University, India. (2) University of Exeter, Penryn Campus, Penryn, TR109FE, United Kingdom.

Cuticular hydrocarbons (CHCs) are key components of the insect cuticle that have contributed to the success and wide distribution of insects. CHCs not only help insects cope with environmental stress but they also help with communication and there can be sex-differences in CHC profiles because of this. Many studies have investigated sex and population differences in CHC profiles, with population studies frequently focusing on latitudinal CHC clines. However, CHC variation across altitudinal clines is less well studied. Here, we test whether CHC profiles change along an altitudinal gradient in the cosmopolitan fruit fly Drosophila melanogaster. We collected three populations from the Western Himalayas along an altitudinal gradient, and characterised their CHC profiles. Additionally, we exposed the flies to desiccating conditions to test for plastic changes in CHC bouquets. We found quantitative differences in male and female across populations, as well as in response to desiccation stress with the overall pattern indicating that long chained CHCs are favoured at higher elevations. However, there were clear patterns of CHC differentiation across altitudes. Thus, the altitudinal data generally recapitulate latitudinal clines.

Feedback control of organ size precision is mediated by apoptosis in the Drosophila eye

22 Mar
09:00-09:30

Saúl Ares
CNB-CSIC

Biological processes are intrinsically noisy and yet, the result of development –like the species-specific size and shape of organs– is usually remarkably precise. This precision suggests the existence of mechanisms of feedback control that ensure that deviations from a target size are minimized. Still, we have very limited understanding of how these mechanisms operate. Here, we investigate the problem of organ size precision using the Drosophila eye. The size of the adult eye depends on the rates at which eye progenitor cells grow and differentiate. We first find that the progenitor net growth rate results from the balance between their proliferation and apoptosis, with this latter contributing to determining both final eye size and its variability. In turn, apoptosis of progenitor cells is hampered by Dpp, a BMP2/4 signaling molecule transiently produced by early differentiating retinal cells. Our genetic and computational experiments show how the status of retinal differentiation is communicated to progenitors through the differentiation-dependent production of Dpp which, by adjusting the rate of apoptosis, exerts a feedback control over the net growth of progenitors to reduce final eye size variability.

The bio-architecture of feather colours

22 Mar
09:30-10:00

Roberto Arbore
CIBIO

Feathers are the most complex epidermal organs of vertebrates. Due to their unique structure, regeneration potential, and enormous variation in shape, colour and function, feathers are powerful models for the study of organogenesis from an evolutionary and developmental perspective. During regeneration, a variety of morphogenetic mechanisms cooccur to build the complex structural organization of feathers which, in turn, forms the scaffold on which colour patterns are formed. The generation of the myriad variations of feather colours and patterns requires the modulation of coordinated cellular behaviours whose molecular bases remain largely unexplored. Using parrots as an example, I will first delineate the general principles of feather morphogenesis, and then I will present our research which, by building on these foundations, aims at advancing our mechanistic understanding of how colour patterns are formed. This knowledge can provide valuable insights on how developmental processes can be finely tuned by natural selection to generate an endless variety of solutions.

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22 Mar
10:00-10:20

Local control of cellular proliferation during organ regeneration in zebrafish

Natalia G. Lavalle(1); Emanuel Cura Costa(1); Jerónimo Miranda-Rodríguez(2); Oriol Viader-Llargués(2); Tomas S. Grigera(1,3,4); Hernan Lopez-Schier(5); Osvaldo Chara(6,7)

(1) Institute of Physics of Liquids and Biological Systems (IFLySIB), National Scientific and Technical Research Council (CONICET), University of La Plata, Argentina; (2) Unit Sensory Biology and Organogenesis, Helmholtz Zentrum München, Munich, Germany; (3) Departamento de Física, Facultad de Ciencias Exactas, Universidad Nacional de La Plata, Argentina; (4) Istituto dei Sistemi Complessi, Consiglio Nazionale delle Ricerche, Rome, Italy; (5) Division of Science, New York University Abu Dhabi, United Arab Emirates; (6) School of Biosciences, University of Nottingham, UK; (7) Instituto de Tecnología, Universidad Argentina de la Empresa (UADE), Ciudad Autónoma de Buenos Aires, Argentina

Regeneration of complex biological tissues is an emergent multicellular behavior during which individual cells with defined phenotypes first detect and then respond to damage through controlled proliferation and differentiation. To understand the mechanism underlying the decision of a cell to re-entry and exit mitotic proliferation we focus on the regenerative neuromasts of zebrafish, and followed a modelling approach. We here investigate this problem by developing a 2D Cellular Potts Model (CPM) that recapitulates the morphological characteristics of regenerating neuromasts and determined the cell-cell affinities to obtain a stable tissue in silico in which mechanisms of cell proliferation depending on local rules are tested. We reproduced the experimental kinetics of sustentacular cells, mantle cells and hair cells constituting the neuromast during laser ablation-derived regeneration by assuming a proliferation switch of sustentacular cells and mantle cells that depends on the number of cells of the same type . Our model predicts that cellular proliferation is a phenotype-independent delayed response to injury, in agreement with previous studies. By comparing model simulations with experimental data, we found that sustentacular cells, the main drivers of regeneration, arrest divisions when surrounded by three neighboring sustentacular cells. However, ancillary mantle cells arrest divisions when confronted by between zero and one neighboring mantle cell. These data indicate that entry and exit of regenerative proliferation are controlled by a local negative feedback loop triggered by the cellular neighborhood.

Modular control of time and space during vertebrate axis segmentation

22 Mar
10:20-10:40

Ali Seleit (1) , Ian Brettell (2) , Tomas Fitzgerald (2) , Carina Vibe (1) , Felix Loosli , Joachim Wittbrodt , Kiyoshi Naruse, Ewan Birney (2)*, Alexander Aulehla (1)*

(1) EMBL, Heidelberg; (2) EBI, UK

How the timing of development is linked to organismal size is a longstanding question. Numerous studies reported a correlation of temporal and spatial traits, yet, the developmental or selective constraints underlying this link remain largely unexplored. We address this question by studying the periodic process of embryonic axis segmentation in-vivo in Oryzias fish. Our interspecies comparison revealed that the timing of segmentation correlated to segment, tissue and organismal size. Segment size in turn scaled according to tissue and organism size. To probe for underlying causes, we genetically hybridised two closely related species. The quantitative analysis in 600 phenotypically diverse F2 embryos revealed a decoupling of timing from size control, while spatial scaling was preserved. Using developmental quantitative trait loci (devQTL) mapping we identified distinct loci linked to either the control of segmentation timing or tissue size. This study demonstrates that a developmental constraint mechanism underlies spatial scaling of axis segmentation, while its spatial and temporal control are dissociable modules.

The cyanobacterial circadian clock couples to pulsatile processes using pulse amplitude modulation

22 Mar
10:40-11:00

Chao Ye (1); Teresa Saez (2); Arijit K. Das(2); James C.W. Locke (2); Philipp Thomas (3); Bruno M.C. Martins (1)

(1) University of Warwick; (2) University of Cambridge; (3) Imperial College London

Circadian clocks generate 24-h rhythms of gene expression in anticipation to daily sunlight cycles. While substantial progress has been made in understanding how cells generate and sustain these rhythms, how clocks are coupled to other cellular networks, what dynamics arise from this coupling, and what biological functions it serves remain open questions. We address these questions using an interdisciplinary approach and the cyanobacterial clock as a model system. Here, I will focus on how the clock interacts with other cyclic processes and regulates the cell division cycle and cellular growth. Bacterial gene expression is in part controlled by sigma factors, which direct RNA polymerase to specific promoters. Through a combination of single-cell microscopy and modelling, we found that the clock modulates the amplitude of expression pulses of the sigma factor RpoD4 that occur at cell division. This pulse amplitude modulation, analogous to AM regulation in radio waves, represents a new class of circadian regulation. It allows the clock to generate robust 24-h periods in RpoD4 expression irrespective of the cellular division rate, even though RpoD4 transcription is pulsatile and tied to cell division. Furthermore, we found that modulating RpoD4 expression allows tuneable control of

cell size in a dose-response manner. While relatively unexplored in gene regulation, pulse amplitude modulation is common in other areas of science and engineering and may be important in other cell types and contexts. Our findings illustrate how simple systems can exhibit complex dynamics, advancing our understanding of the interdependency between gene circuits and cellular physiology.

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22 Mar
11:30-12:00

Are there "universal" laws of cellular growth?

Marco Cosentino Lagomarsino
IFOM/U. Milan

Proliferating cells generally organize their resources in order to harness nutrients from the environment and grow. Work in bacteria has highlighted how this behavior leads to emergent "growth laws" linking growth to cellular composition. However, we still have limited insight on the generality of such laws and the mechanisms underlying them. After reviewing some key general aspects of cellular growth, I will address this question through two specific findings. First, while ribosome autocatalysis sets the growth rate, total mRNA can affect growth by determining the amount of free ribosomes. Second, coupling of nutrient sensing and transcriptional regulation of ribosome biogenesis naturally leads to oscillatory behavior.

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22 Mar
12:00-12:20

Molecular sorting on a fluctuating membrane

Andreghetti, Damiano (1); Dall'Asta, Luca (1,3,4,5); Gamba, Andrea (1,3,4); Kolokolov, Igor (2,6); Lebedev, Vladimir (2,6)

(1) Politecnico di Torino; (2) L.D. Landau Institute for Theoretical Physics; (3) Italian Institute for Genomic Medicine; (4) Istituto Nazionale di Fisica Nucleare; (5) Collegio Carlo Alberto; (6) National Research University Higher School of Economics

Molecular sorting is a fundamental ordering process taking place in eukaryotic cells. In this process, submicrometric lipid vesicles enriched in specific biomolecules are distilled, thus countering the homogenizing effect of diffusion. Molecules attach to lipid membranes and diffuse laterally. Due to a variety of short- and long-range interactions, the molecules tend to aggregate in domains characterized by specific chemical compositions. Then, domain formation promotes membrane bending and fission, leading to the engulfment of the formed domains into separate lipid vesicles. The enriched vesicles are finally delivered to the appropriate intracellular destinations by means of molecular motors. This distillation process can be investigated at the mesoscopic scale from a statistical physics perspective. Molecular distillation is a driven nonequilibrium process, that reaches optimal efficiency for intermediate values of the driving force. Along with intermolecular contact interactions, it is known that membranes fluctuations can mediate longer-range interactions between biomolecular inclusions exhibiting a larger than average bending rigidity. Our study focuses on the role of these fluctuation-induced interactions in the molecular distillation process. By means of numerical simulations we show that fluctuation-induced forces favor distillation in the parameter region where contact interactions are weak, and impair it in the region of strong contact interactions.

From structural variability to protein energetics of a molecular motor

22 Mar
12:20-12:40

Mello, V H (1); Wald J (2); Marlovits T (2); Sartori, P (1)

(1) Instituto Gulbenkian de Ciência; (2) Centre for Structural Systems Biology

Molecular motors harness chemical energy from molecules, such as ATP, to perform mechanical work. Through this process, energy is transiently stored in deformations of the protein structure. However, the assessment of these deformations at an atomic scale is often based on visual comparisons of resolved structures. Here we apply the formalism of elasticity theory to quantify deformation in protein atomic models. In particular, we use this approach for understanding how molecular motors perform mechanochemical energy transduction. We choose RuvB as a model system, a molecular motor crucial for branch migration in bacterial homologous recombination. Importantly, multiple cryo-EM conformations are available for different substeps of the catalytic cycle. We compute the spatial distribution of elastic energy across the protein structure throughout the mechanochemical cycle. For each residue, we obtain a 30-states energy trajectory, spanning a range of four orders of magnitude and revealing an intricate spatiotemporal energy redistribution. Analysing this data, we identify outlier residues on the energy distribution, which constitute regions of interest that are mechanically active in different sets of conformations. These regions link structure and energy transduction in protein-protein and protein-nucleotide interactions. Finally, we build a stochastic model consisting of ATPases coupled in a ring which is constrained by the energetics inferred from the structural data. Our kinetic model is consistent with single-molecule biophysical experiments and highlights the significance of subunit cooperativity for sequential dynamics. In summary, our methodology provides a physical interpretation of structural data, bridging detailed atomic-scale information and molecule-scale modelling.

Using pseudo-embryos to study the Hox timer

22 Mar
14:00-15:00

Hocine Rekaik (1), Lucille Lopez-Delisle (1), Bénédicte Mascrez (2),Alexandre Mayran (1) and Denis Duboule (1,2,3)

(1) School of Life Sciences, Ecole Polytechnique Fédérale de Lausanne (EPFL), Switzerland; (2) Department of Genetics and Evolution, University of Geneva, Switzerland; (3) Collège de France, Paris, France

During vertebrate development, clustered Hox genes are activated in a precise time-sequence, leading to patterns necessary to properly establish the body plan. The mechanism underlying this phenomenon (the Hox timer) has remained elusive ever since its initial observation in 1989, due to the difficulty to approach it using early gastrulating mouse embryos. I will discuss our recent results using stembryos produced out of ES cells ('gastruloids') as an alternative approach to address this question and will show that the temporal dynamic of the system may rely upon the use of series of CTCF sites as successive boundary elements. While this mechanism can secure the deployment of Hox gene transcription and hence the proper establishment

of axial structures within any given vertebrate species, it also offers some evolutionary flexibility, for minimal modifications in the number, position or affinity of these sites would translate into heterochronic transcription. The relationship between the topology of these binding sites and a segmented body plan will then be discussed.

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22 Mar
15:00-15:30

1993 - 2023: 30 Years of Cytokine Signaling

Dimitrios Stavropodis
U. of Athens

Cytokines are a broad category of small proteins that regulate diverse functions of multiple lineages, via cell signaling engagement. A typical cytokine binds onto its cognate transmembrane receptor and transmits a variety of signals downstream. Strong and high affinity binding of a cytokine to its receptor causes trans-phosphorylation and subsequent activation of one or more Jak family members (Jak1, Jak2, Jak3 and Tyk2). A Jak tyrosine kinase, in a phosphorylated and activated state, can, then, phosphorylate and activate one or more of the Stat family members (Stat1, Stat2, Stat3, Stat4, Stat5a, Stat5b and Stat6), on a critical tyrosine. A typical Stat transcription factor, in its tyrosine-phosphorylated and activated form, can, next, (homo-)dimerize, enter the nucleus, and bind onto promoter sequences of target genes to activate their transcription. Cellular transgenesis and gene knock-out technologies prove the major role of Jak/Stat signaling in a plethora of biological processes and responses, including -among others- the ones being induced by erythropoietin, prolactin, growth hormone, interleukins (e.g., IL-2 and IL-7) and interferons (e.g., IFN-gamma). Profiling of the functional/active(ated) kinome, through employment of the state-of-the-art PamGene (PamStation12/PamChip/BioNavigator) technology, during development, tissue patterning and cell pathology, holds strong promise for the chemical drugging of abnormal kinases and targeted therapy of human diseases, including cancer.

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22 Mar
15:30-15:50

Correspondence between multiple signaling and developmental cellular patterns: a computational perspective

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The spatial arrangement of variant phenotypes during stem cell division plays a crucial role in the self-organization of cell tissues. The patterns observed in these cellular assemblies, where multiple phenotypes vie for space and resources, are largely influenced by a mixture of different diffusible chemical signals. This complex process is carried out within a chronological framework of interplaying intracellular and intercellular events. This includes receiving external stimulants- whether secreted by

other individuals or provided by the environment- interpreting these environmental signals and incorporating the information to designate cell fate. An enhanced understanding of the building blocks of this framework would help to set the scene for promising regenerative therapies. In this study, by proposing a designative computational map, we show that there is a correspondence between multiple signaling and developmental cellular patterns. That is, the model provides an appropriate prediction for the final structure of the differentiated cells in a multi-signal, multi-cell environment. Besides, given that the final state of the cellular organization is known, the corresponding regressive signaling patterns are partly predictable following the proposed map.

Observing DNA damage responses of cells with Data-Driven Microscopy

22 Mar
15:50-16:10

Subburam, Kesavan (1); Nordenfelt, Pontus (1)

(1) Faculty of Medicine, Lund University

The genetic information stored in the DNA is subject to stochastic and continuous changes from internal and external factors in the cells. To preserve the stability of their genome, cells have evolved a host of mechanisms that detect and reverse damage to the DNA. Such mechanisms operate under the constraints of the overall cellular structure and processes. Understanding the responses of the different subsystems of the cell under damage is crucial to realising the overall framework of the organisation of life at the microscopic level. Traditionally, DNA damage responses (DDR) have been studied extensively at the protein and pathway level with ensemble biochemistry and molecular biology tools. However, such ensemble measurements have limited readout resolution, and do not capture the heterogeneity in response and spatiotemporal relationships between cellular subsystems. With the development of high throughput microscopy-based assays, it has become possible to investigate the relationship between the different factors involved in DNA damage responses and the constraints imposed by cellular organisation at different length and time scales on a cell-by-cell basis. Further, by developing dynamic control of the microscope with advanced data-driven software architecture has allowed for incorporating additional context onto events and extracting extra information from cells under observation. In this work, we demonstrate generalizable applications of data-driven microscopy for studying DNA damage responses in live and fixed cells to map the cell-cycle stage and correlate it with heterogeneity in damage response with readout from live and immunofluorescence imaging of various factors of repair. We believe this method can be applied in diverse contexts of DDR to paint a larger picture of overall cellular organisation.

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22 Mar
16:40-17:00

Understanding mitotic physiology of pluripotent stem cells

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Pluripotent stem cells (PSCs) hold great promise in basic and translational research. However, in culture these cells present genomic instability that can lead to aneuploidy, compromising further applications. The molecular mechanisms underlying this genomic instability remain poorly described. Here, we explore possible reasons for compromised mitotic segregation, building on known differences in the PSCs' chromatin state. We have previously shown that PSCs have weaker centromeres. Comparative analysis of PSCs and their somatic counterparts' mitotic defects revealed a high frequency of lagging chromosomes in PSCs. These findings suggest that defects in centromere/kinetochore assembly, and consequently chromosome attachments may be a major mechanism underlying the observed high frequency of chromosome segregation errors. In addition to changes in the centromeres, PSCs have less compacted DNA during interphase, and we observed a high frequency of condensation alterations during mitosis. However, we did not observe a high frequency of DNA bridges, indicating that the anticipated problems in chromosome condensation and/or resolution of chromosome entanglements are efficiently resolved. These findings suggest that PSCs may have compensatory mechanisms to overcome the less compact state. To explore this possibility, we have analysed levels of key proteins involved in mitotic chromosome organization, the condensin complexes. We observed that the chromatin bound levels of Condensin I are increased in PSCs when compared to somatic cells. Ongoing work aims to how the excess of Condensin I can work as a putative compensation mechanism to boost PSCs' mitotic fidelity. Understanding PSCs mitotic´s physiology will open questions to better comprehend mitosis during development.

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22 Mar
17:00-17:20

Interactions across scales: protein-erythrocyte and erythrocyte-erythrocyte binding as determinants for cardiovascular risk

Carvalho, Filomena A.; Lopes, Catarina S.; Santos, Nuno C.

Instituto de Medicina Molecular, Faculdade de Medicina, Universidade de Lisboa, Lisbon, Portugal

Erythrocytes are deformable cells that undergo progressive biophysical and biochemical changes affecting normal blood flow. Fibrinogen, one of the most abundant blood plasma proteins, is a primary determinant for changes in hemorheological properties, and a major independent risk factor for cardiovascular diseases [1]. We have previously demonstrated the biomedical relevance of the measurement of fibrinogen-erythrocyte and erythrocyte-erythrocyte interactions, using atomic force microscopy (AFM)-based force spectroscopy, at the level of clinical prognosis in

heart failure and essential arterial hypertension patients [2-4]. Recently, the adhesion between human erythrocytes was further assessed by comparing AFM-based force spectroscopy measurements with micropipette aspiration technique, in the absence and presence of fibrinogen [5]. These experimental data were then used in the development of a mathematical model to examine the biomedical relevant interaction between two erythrocytes [5]. More recently, we evaluated changes in fibrinogen-erythrocyte and erythrocyte-erythrocyte interactions in carotid artery disease (CAD) patients, and characterized the biomechanical properties of carotid atherosclerotic plaques from CAD patients. Blood samples collected from CAD patients, before and after endarterectomy surgery, were analyzed and compared to the control group. These studies comprised hemorheological parameters and AFM measurements of fibrinogen-erythrocyte and erythrocyte-erythrocyte interactions, and cell elasticity.

Abstracts - Posters

Intragenic ohnolog miRNAs 1

Agasso, Leonardo(1); Molineris, Ivan(2); Caselle, Michele(1)

(1)University of Turin - Physics Department and INFN - Section of Turin; (2) Department of Life Science and System Biology, University of Turin

We worked on the identification and analysis of intragenic miRNA pairs in the human genome derived from the two rounds of Whole Genome Duplication (WGD) occurred in the early vertebrate lineage. The analysis focuses on their role in the structure of the human gene regulatory network. We retrieved such pairs by looking for miRNA pairs harbored on protein-coding ohnolog (i.e., duplicates originating from WGD events) gene pairs; these putative intragenic miRNA pairs show a stronger tendency to be retained when harbored on ohnolog gene pairs compared to analogous miRNA pairs harbored on gene pairs originating from Small Scale Duplication (SSD) events. This observation is supported by the evidence for a stronger tendency of putative intragenic ohnolog miRNA pairs to regulate common target genes (relying on different available databases) and have a better preserved mature sequence. We argue that these particular characteristics can be explained in terms of dosage balancing and stoichiometric balance. Analyzing the role of putative intragenic ohno-miRNAs in the human gene regulatory networks we're currently aiming to further assess some preliminary results showing that putative intragenic ohno-miRNAs are also statistically overrepresented in specific network motif whose functional significance is widely acknowledged (e.g. bifan motif).

Deciphering Cellular Crosstalk with Niche Covariation: Insights from Single-Cell Spatial Transcriptomics Data 2

Agrawal Ankit (1), Stefan Thomann (1), Dominic Grun (1)

(1) Würzburg Institute of Systems Immunology, Max-Planck Research Group at the Julius-Maximilians-Universität Würzburg, Germany

Cellular states are characterized by distinct molecular configurations that determine fate and function. Within tissues, cell state variability can be divided into intrinsic and extrinsic determinants. Both intrinsic factors, including noise in gene expression and external influences from the microenvironment, shape the subtleties of gene expression variation within specific cell types. Distinguishing between intrinsic and extrinsic elements of cell state variability is essential for comprehensively

understanding the nature of cell state transitions and deciphering the mechanisms underlying tissue homeostasis. Our study introduces niche covariation (NiCo) that constructs a classifier, based on the spatial neighborhood colocalization of the cellular state as a function of extrinsic components to understand the cell type identity. Furthermore, Nico infers spatial covariation of latent factors that capture cell state variability in tissue niches and interprets these factors by leveraging transcriptome-wide information from the reference data. Subsequently, regression uncovers linear dependencies between latent spatial factors niche to unveil gene module interactions. Ligand-receptor and pathway enrichment analysis was used to interpret gene involvement in these covarying modules. By applying NiCo to the developing mouse embryo, small intestine, and liver, we predict novel niche interactions underlying cell state variation. In particular, NiCo predicts a feedback mechanism between Kupffer cells and neighboring stellate cells, limiting stellate cell activation in the normal liver. NiCo's precision in deciphering complex cellular dialogues provides a new lens through which we can explore the complex language of cellular communication, promising deeper insights into tissue dynamics and pathology.

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3 Approaching the mechanistic basis of Disease tolerance in Drosophila melanogaster

P.A. Akyaw1, D. Roque1, 2, T.F. Paulo1, D. Duneau3, E. Lafuente1, É. Sucena1, 2

1. Instituto Gulbenkian de Ciência, Oeiras, Portugal,2. Departamento de Biologia Animal, Faculdade de Ciências da Universidade de Lisboa, Lisboa, Portugal,3.Institute of Evolutionary Biology, University of Edinburgh, Scotland, United Kingdom.

Immune response against infections can be divided into mechanisms of Resistance that ensure active pathogen elimination, and of Disease Tolerance, which include host-tissue protection and repair processes that will return the host to physiological homeostasis. Studies trying to understand host responses to infection have mostly targeted mechanisms of Resistance, and consequently, these are now well described in both vertebrates and invertebrates. However, the mechanistic basis of Disease Tolerance is poorly understood, in part because both mechanisms interact and are difficult separate. To tease apart these components, we have developed a protocol of oral exposure of Drosophila melanogaster to inactivated Pseudomonas entomophila, hence, minimizing the Resistance component of the host response. To get to the mechanisms underlying the variation in host Disease Tolerance, we have applied this protocol to 200 Drosophila melanogaster isogenic lines (DGRP) and measured fitness traits (survival and reproduction). Using this protocol, we observed considerable host genetic variation for these traits that show weak correlation with one another. The observed host variation is highly sexually dimorphic and differs according to the level of pathogenicity of the bacterium species to which it was exposed. In addition, we will be presenting a preliminary account of a genome-wide association study on these lines, for these traits, that includes functional validation of candidate genes and the first glimpse at the genetic bases for Disease Tolerance mechanisms. Furthermore, we will compare these data with two more datasets, one using exposure to non-virulent strains and, the other, utilising host mutants incapable of mounting

immune-responses. With this comparison we expect to partition the effects and mechanisms of Disease Tolerance into its immunopathology and infection-induced damage components.

Exploring Spatial Segregation Induced by Competition Avoidance as Driving Mechanism for Emergent Coexistence in Microbial Communities 4

Mattei, Mattia; Arenas, Alex

U. Rovira i Virgili

We develop an individual-based spatial simulation to depict the individual movement of bacteria, leading to the formation of spatially segregated clusters resulting from the escape from regions with high competition. This simulation is then utilized to calculate the initial conditions for a metapopulations Lotka-Volterra model with only competitive interactions, capturing the growth dynamics of such patches of bacteria. This study shows that: i) segregation of clusters of bacteria can be obtained as a result of competition avoidance only and, therefore, it can potentially occur in any conditions regardless of the environmental setup; ii) considering a metapopulation structure in a Lotka-Volterra formalism alters considerably the pattern of coexistence for the species and iii) spatial segregation and bacterial self-organization can be seen as the driving mechanisms behind the formation of stable coexisting small microbial communities, the majority of which do not coexist when isolated in pairwise combinations.

High Performance quantitative image analysis: a S. aureus case study 5

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"Fluorescence microscopy, coupled with powerful quantitative image analysis methods, has played a pivotal role in unveiling biological processes at the single cell level. Simultaneously, the expanding scale and complexity of microscopy datasets requires accelerated analytical workflows. Our new Python image analysis package,

NanoPyx, meets this need through an adaptive framework enhanced for high-speed bioimage analysis. At the core of NanoPyx, the Liquid Engine dynamically generates optimized CPU and GPU-based code variations, learning and predicting the fastest implementation based on each individual user's input data and hardware. This data-driven optimization achieves considerably faster processing, becoming broadly relevant to reactive microscopy and computing fields requiring efficiency, and contributing to advances in tackling important human health issues. One example of an important human health issue where quantitative image analysis can bring added value is antibiotic-resistant infections, which are frequently caused by methicillin-resistant Staphylococcus aureus (MRSA). As the cell wall is essential for bacterial viability, proteins involved in its synthesis and regulation are prominent targets for antibiotics which makes the study of peptidoglycan (PG) synthases particularly interesting. The exact mechanisms of regulation and the dynamics of PG synthases inside a living S. aureus cell is still poorly understood. In this work, using Single Particle Tracking (SPT) microscopy, we show that we can infer the three-dimensional trajectories of single PG synthases from two-dimensional images. Using an integrated approach to the analysis of protein trajectories within S. aureus, we propose a model where a single population of moving PG synthases drives constriction during division."

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The intrinsic dimension of cell differentiation

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Despite the striking simplicity of the idea behind the Waddington Landscape, the identification of a quantity that approximates the differentiation potential is still an open area of research. Our goal is to identify a proxy for this differentiation potential avoiding biological assumptions, strong pre-processing and sophisticated approaches, focusing instead on the statistical and geometrical properties of scRNA-seq datasets. The mRNA count matrix contains the coordinates of N cells in the high-dimensional expression space, where the dimensions are given by the sequenced genes. However, not all these directions can be freely explored by cells, because of biological constraints and correlations between genes: geometrically, this simple hypothesis implies that the data points are not uniformly spread along each direction, but are rather embedded in a subspace of dimension lower than that of the original expression space: to prove it, we are focusing on the intrinsic dimension, which can be interpreted as the dimension of the manifold from which the data are supposed to be drawn. The second conjecture we want to prove is that the intrinsic dimension reflects the level of stemness of a cell population. To investigate these insights, we exploit standard intrinsic dimension estimators that have been developed in the context of data science, based on principal component analysis (global) and two nearest neighbors (local). We studied several datasets referring to different biological processes and animal models, and we observed that the intrinsic dimensionality

decreases during specialization, justified by a progressive structuring of data due to gene regulation mechanisms.

Common and distinct activities of EMT factors of the ZEB family in glioblastoma cancer stem cells 7

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Glioblastoma is the most lethal brain tumor in adults, due to its highly invasive nature and the presence of glioblastoma stem cells (GSCs). We recently showed that the classical Epithelial-to-Mesenchymal-Transition (EMT) transcription factor ZEB1 regulates an EMT-like program that contributes to the high invasive capacities of GSCs. ZEB1 simultaneously promotes gene activation and repression at a genome-wide scale. Repression is associated with the direct binding of ZEB1 to regulatory regions of "epithelial genes", while indirect recruitment, via LEF/TCF factors, leads to transcriptional activation. To which extent EMT-TFs regulate common versus unique functions is an outstanding question in EMT field. We aimed at addressing this, by providing a comprehensive view of ZEB factors in glioblastoma. ChIP-seq characterization of ZEB1 and ZEB2 binding events revealed identical binding profiles at gene regulatory regions, defined by characteristic chromatin states. This includes binding events resulting from both direct and indirect modes of recruitment. Strikingly, while both factors promote transcriptional repression via direct binding to target genes, they display antagonistic activities (ZEB1 activation/ZEB2 repression) when recruited by LEF/TCF factors. Functional studies indicate that such differences cannot be solely attributed to an N-terminal domain unique to ZEB1, which mediates recruitment of the co-activator p300. Altogether, we propose a model whereby ZEB factors regulate partially overlapping, partially distinct, EMT-like programs in glioblastoma. We speculate this may be associated with the adaptation of GSCs to tumor microenvironment. This is in line with preliminary analysis, showing differences in ZEB1 and ZEB2 expression across distinct tumor niches in glioblastoma patient samples.

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Discovering the temporal footprint of phototoxicity

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Live-cell fluorescent microscopy is widely used to observe the living matter from nano to macro scales. However, the underestimated effects of phototoxicity in fluorescence imaging, hinders our ability to accurately study the natural behaviour of biological systems and can impact experimental reproducibility. In this work, we propose a novel approach, Phototoxicity Fitness Time Trial (PhotoFiTT), to quantify phototoxicity effects in live-cell fluorescence microscopy. PhotoFiTT utilises deviations from known sequential cellular events to predict cellular photodamage without the need for fluorescence labelling. It measures light-induced abnormal cellular dynamics and delays in cell cycle progression after light irradiation using machine learning and analytical techniques. PhotoFiTT reveals a measurable relationship between light dose, wavelength, and phototoxicity, which characterises progressively worsening cell photodamage. The characterisation of phototoxicity at a local and cellular level is essential for further understanding of photodamage in more complex systems such as tissues and embryo development. We anticipate that PhotoFiTT will contribute to the foundations of improved imaging pipelines that target sample health, enabling the preservation of sample physiological conditions and behaviour, and expanding the applicability of live-cell fluorescence microscopy.

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@IGCHistopath

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Instituto Gulbenkian de Ciência

The Histopathology Facility (HF) is a user-oriented, fully equipped, laboratory that provides high-quality histopathological services and pathology support both to the Instituto Gulbenkian de Ciência (IGC) scientific community and to external users (associate laboratories, academic institutions, and industry). The facility staff is highly qualified and has ample experience working with a diverse range of samples: from human to various model organisms and scaffold membrane, spheroid and organoid cultures. However, modern-day Histopathology is more than the branch of biology that studies the microscopic anatomy and pathology of biological tissues. In biology, much like in real estate, location matters, and contemporary histology techniques have grown to bridge the gaps between different scales to provide wider,

stronger and more detailed data to answer biological questions. Accordingly, since 2016, and in close collaboration with other facilities in the institute, HF has been implementing techniques such as Correlative Light-Electron Microscopy (CLEM), Stereology, and both Formalin Fixed Paraffin Embedded and Fresh Frozen Spatial Transcriptomics.

Evolutionary repairing anaphase

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Advances in comparative genomics are demonstrating remarkable biodiversity in how different organisms perform important cellular functions. Notably, the discovery of organisms lacking genes conventionally deemed essential for viability has challenged our established comprehension of ubiquitous biological processes. For instance, Giardia lamblia has been found to undergo cell division without an Anaphase-Promoting Complex (APC), a ubiquitin ligase which is essential to promote mitotic progression in all known eukaryotes. This raises the question of how an organism can lose essential genes while maintaining viability. To address this point, we generated temperature-sensitive mutants of two APC subunits (doc1 and cdc26) through genetic manipulation of the S. cerevisiae genome. We subjected single and double mutants to laboratory evolution for approximately 250 generations with gradual temperature increase from 23 to 37^{o}C. While ancestor mutants die with a terminal mitotic arrest at 37^{o}C, due to the inactivation of the APC complex, evolved clones retained mitotic proficiency at the final temperature. Phenotypic characterization of the strains, combined with whole genome sequencing will reveal alternative strategies that cells can use to undergo cell division once the more conserved mechanisms are perturbed, and inform on how organisms can significantly change core aspects of their cell biology and the strategies they may adopt to do so.

To stress... to resist? The power of stress responses in sepsis

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Restoration of organismal homeostasis after an insult is the result of a balanced response by the two main arms of host defence: disease tolerance and resistance. Inducing stress responses can trigger disease tolerance mechanisms, as demonstrated by our lab (DOI: 10.1016/j.immuni.2020.09.011). In a severe murine sepsis model we showed increased survival and less tissue damage upon treatment with drugs that

induce mild mitochondrial stress. However, the protective mechanisms initiated by stress responses have not been systematically studied. This work focuses on understanding the impact of stress responses on resistance and tolerance mechanisms. For this we optimized a mild murine sepsis model to allow monitoring for 5 days. Within this period, pathogen load, immune phenotyping and damage markers were accessed. Mixed effects models, with experiments as random effect, were performed. Data was analysed using R. So far, we have gathered evidence showing a significant pathogen load decrease in treated animals starting at 72h post-infection and treatment. This shows that mild mitochondrial stress is also triggering pathogen clearance, a hallmark of resistance responses. Furthermore, we found significant differences in specific immune cell populations associated with the treatment of septic animals, which could be core players on the underlying protective mechanisms. The results of this work, show that mild mitochondrial stress can improve resistance mechanisms, in addition to tolerance in septic mice. Understanding the mechanistic bases by which these stress responses are restoring homeostasis could contribute to the development of synergistic therapeutic approaches to life-threatening conditions characterized by dysfunctional immune responses.

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12 DL4MicEverywhere: Deep learning for microscopy made flexible, shareable, and reproducible

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Contextualising and understanding biological phenomena rely on the ability to extract meaningful information at different scales and relate it across dimensions. While life can be observed from the macro to the microscale, even with molecular specificity, simultaneous observation of the living matter at different scales remains a challenge. Recently, deep learning-based imaging techniques have pushed the limits of multimodal high-throughput high-content imaging. These advanced techniques can virtually super-resolve extended fields of view, enable increased acquisition times with minimized photodamage, or identify targets for unbiased acquisitions in data driven microscopy, thus, providing us a broader context of the sample in space and

time, and having a better understanding of its natural behaviour. Nevertheless, the complexity and expertise required for these methods prevents biologists with limited coding ability from exploiting them. Moreover, deep learning-based analysis depends on specific programmatic packages and versions which are constantly updated, posing a major reproducibility issue. To address this limitation, we developed DL4MicEverywhere, an easy-to-use tool to pack and freeze the environments containing all the dependencies required for the deployment of deep learning techniques. DL4MicEverywhere offers a collection of user-friendly notebooks for segmentation, super-resolution or artificial staining, encapsulated in Docker containers compatible with different operating systems, ensuring the long-term functionality and reusability of the methods. DL4MicEverywhere aims to support image driven research by bringing long standing solutions suitable for scientific projects development timings, as well as enabling the scalability of next generation image data analysis.

Coupling the developmental genetics of locomotion and mating structures

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The hindlimb of tetrapods and the external genitalia of the amniote lineage are thought to derive from an ancestral common primordium that evolved to generate a wide diversity of structures, adapted for efficient locomotion and mating. During embryo development, these structures are developed in proximity from different tissues, in the posterior end of the trunk during the trunk-to-tail transition. Mechanistically, the hindlimbs and the external genitalia share common genetic regulatory factors. It has been shown that both the trunk-to-tail transition and the induction of these appendages are controlled by the Tgfbr1 signaling pathway, whereas their commonalities regarding their developmental origin are poorly understood. To explore how these two appendages are developed, we used the Tgfbr1 conditional knock out (Tgfbr1-cKO) mouse embryos, revealing a phenotype of duplication of the hindlimbs and underdevelopment of the external genitalia. Following genetic characterization of the embryonic primordia, we reasoned that the progenitor tissue of the external genitalia can change its fate towards hindlimbs. Characterization of the chromatin accessibility profiles from these tissues reveals that Tgfbr1 controls the response of the progenitor tissues to common key regulators. To understand the transcriptional circuit of both appendages, as well as the fate decisions of progenitor cell populations of the genital tissue, we are currently carrying out a single cell time-course transcriptomic approach. Our work uncovers a remarkable tissue plasticity with potential implications in the evolution of the hindlimb/genital area of tetrapods and identifies a novel mechanism for Tgfbr1 activity that might also contribute to the control of other physiological or pathological processes.

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14 Hybrid structural arrangements mediate stability and feasibility in mutualistic networks

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Perhaps the largest debate in network Ecology, the emergence of structural patterns stands out as a multifaceted problem. To the methodological challenges – pattern identification, statistical significance – one has to add the relationship between candidate architectures and dynamical performance. In the case of mutualistic communities, the debate revolves mostly around two structural arrangements (nestedness and modularity) and two requirements for persistence, namely feasibility and stability. So far, it is clear that the former is strongly related to nestedness, while the latter is enhanced in modular systems. Adding to this, it has recently become clear that nestedness and modularity are antagonistic patterns – or, at the very least, their coexistence in a single system is problematic. In this context, this work addresses the role of the interaction architecture in the emergence and maintenance of both properties, introducing the idea of hybrid architectural configurations. Specifically, we examine in-block nestedness, compound by disjoint subsets of species (modules) with internal nested organization, and prove that it grants a balanced trade-off between stability and feasibility. Remarkably, we analyze a large amount of empirical communities and find that a relevant fraction of them exhibits a marked in-block nested structure. We elaborate on the implications of these results, arguing that they provide new insights about the key properties ruling community assembly.

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15 Bioimaging across scales

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Biological imaging technologies play a pivotal role in advancing our understanding of living organisms at the molecular, cell, tissue, organ and organism levels. BioImaging tools ranging from microscopy to medical and pre-clinical imaging modalities, give life-scientists and medical professionals the possibility to study cellular structures, biological processes, track organism and disease development and progression. Applications extend across numerous fields including biology, development, neuroscience, physiology, medicine, providing valuable insights for fundamental research, diagnostics, treatment development. In this presentation I will provide a quick overview of several of state-of-the-art bioimaging technologies currently available, and how they are being used to address several different biological questions at the various biological scales.

Live-cell microscopy: less is more

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The midbody remnant is a structure that is released into the extracellular environment during cell division and can be subsequently reabsorbed back by a cell and play a role in cell proliferation. However, the molecular mechanisms and dynamics of the reabsorption process are still poorly understood. As a rare and fast event, it is very challenging to image with conventional fluorescence microscopy. Indeed, a high imaging speed is required, but maintaining it for long periods of time will induce phototoxic effects and alter the cell's normal behaviour. This also limits the number of cells that can be monitored and, thus, the number of observations. To increase efficiency, we are developing a data-driven microscope that can analyse the images acquired in real-time and adjust the acquisition parameters targeting this event. In our implementation, the microscope can identify the cell's cycle and midbody stages and, according to this, decide where, when and how to image. To achieve this goal, we are working with a genetically modified cell line where the nucleus expresses different fluorescent proteins depending on the cell cycle stage, allowing us to distinguish them. We are using deep learning methods to generate these fluorescent-like images from brightfield images. Thus, we can monitor the cell cycle and identify specific cell stages with brightfield microscopy, which is more live-cell compatible. Finally, we also show an example of triggered acquisition, where the microscope identifies mitotic cells with widefield microscopy and then acquires them with super-resolution microscopy.

Thymus formation in uncharted embryonic territories

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The thymus is a conserved organ among vertebrates, derived from the endoderm of distinct pharyngeal pouches (PP), whose location and number vary across species. Together with reports of sporadic ectopic thymus locations in mice and humans, this suggests that the potential to make a thymus resides in a broader region of the PP endoderm than previously ascribed. Using the chick-quail chimera system, we explore this hypothesis and test the capacity of non-canonical pouches to participate in thymus formation. We further ask if the local mesenchyme of pharyngeal arches (PA) could also play a role in the regulation of thymus formation. After testing several embryonic tissue associations, we mapped the pharyngeal endoderm

regions with thymus potential to the second and third/fourth pharyngeal pouches (2PP and 3/4PP). We further identified mesenchyme regions that regulate this potential to the 3/4 pharyngeal arches and to the dorsal region of the second arch, with positive and negative influences, respectively. Transcriptomic analysis of these tissues helped us revealing a common genetic program in the PP endoderm linked to thymus potential in addition to finding distinct signalling pathways involved in the cellular interactions with the mesenchyme of the pharyngeal arches that result in modulating this potential. Together, these results provide new information about the initial specification of thymus primordia in the embryo that may contribute to improving the development of thymus organoid systems.

From the molecule to a new mouse model: a tour of IGC Mouse Transgenics Unit (MTU)

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At the IGC MTU we tailor your mouse model! And it starts with a drawing on a paper, where ideas for the desired genetic alteration are sketched, while discussing the possibilities, pros and cons and more importantly, the feasibility of the project. Not all the good ideas that come up to create an animal model can be transferred to the DNA molecule, even using the ultimate state of the art technologies in the field, but with the CRISPR-associated protein 9 (Cas9) system for gene editing, we can say, almost all of it can! Our custom made genetically modified mice can also be done via pronuclear microinjection of conventional expression constructs and Bacterial Artificial Chromosomes (BACs) or injecting gene-targeted ES cells into host blastocysts to generate gene knock-out or knock-in mice. So, after deciding on how to do it, i. e., after choosing from the possible methods we have to engineer the genetic information of the mouse, it starts the design at the level of the DNA molecule in silico. Our tour then guides us to the lab, where we build (if not ordered) chemical and biological tools, at the RNA, DNA and protein levels. The mouse work starts in a dedicated area inside the Institute, where, in a very controlled environment, we micromanipulate mouse embryos at very early stages of development, to have the so desired new mouse to your research.

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gamma-Tubulin controls primary cilia length

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Gamma-Tubulin is one of the five Tubulins present in humans, and it is very well-known for its core function in nucleating microtubules as part of a bigger complex called the "ring-complex". Although most reports relate g-Tubulin function with that of the main tubulin organising centre (MTOC) in dividing cells, the centrosome, few others pay attention to its function at the MTOC in G0 cells, the cilia base or basal body (BB).

Primary cilia extend from the cell surface of most mammalian cells, playing essential roles in sensory function and cell signalling, among others. Thus, their dysfunction is linked to several rather heterogeneous and thus debilitating disorders. Despite the broad knowledge on ciliogenesis and the role of g-Tubulin in this specific but short process, little is known about g-Tubulin role during the maintenance of the cilia at the cell surface.

Our recent studies in human cells show that knocking down g-tubulin leads to longer, thinner and irregular cilia. In addition, these cilia show impairment in either localisation and/or amounts of key structural proteins normally placed at the BB and the axoneme during cilia maintenance.

These preliminary results suggest: 1) cilia maintenance needs an active and continuous regulation; 2) g-Tubulin has a role in this cilia maintenance regulation, specifically in controlling cilia length, that is beyond its previous known functions.

Further research will explain the biological relevance of this role and the molecular mechanism of g-Tubulin in cilia maintenance regulation.

Artificial selection for sociality changes the brain transcriptome in zebrafish

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Marta Liber, Susana A. M. Varela, Magda Teles, Jingtao Lilue, Rui Oliveira

IGC

It is possible to observe similar behavioral phenotypes like pair bonding or maternal care, across diverse animal species. This leads to the question of whether shared behaviors have a common genetic basis - behavioral toolkit, that is conserved and reused throughout evolution to embed similar behaviors. Studies suggest that behavioral phenotypes are associated with changes in expression of multiple genes across different brain regions, creating regulatory networks at molecular and functional levels. In this work we computationally explore the genes and gene regulatory networks underlying the evolution of sociality. An artificial selection experiment for high and low sociality lines in zebrafish has been ongoing in the lab, using a social preference paradigm (i.e., video of a mixed-sex conspecific shoal vs video of moving circles), and the individual preference to associate with a shoal or the circles was quantified. After 3 generations, the lines selected for sociality started to diverge significantly in social preference from the other lines (circle and control). The

transcriptomic profiles of these three lines were analyzed in different brain regions, and the results suggest the presence of differentially expressed genes associated to each selected line. Further, a Weighted Correlation Networks Analysis (WGCNA) has been applied, which allowed us to find clusters of genes specific to each line as well. Considering that a social behavior can be regulated not by a particular gene, but also by a set of molecular pathways, our next step is to search for conserved mechanisms underlying sociality, across neuronal networks, at single-cell resolution.

Enabling Cell Contraction: Endoplasmic Reticulum-Plasma Membrane Contact Sites are established by Arp2/3 complex for contraction of Skeletal Muscle

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Skeletal muscle plays a vital role in body movement by generating force through cell contraction. Contraction starts at the motor neuron, which induces muscle cell (myofiber) membrane depolarization leading to calcium release from the endoplasmic reticulum (ER), causing the myofiber to contract. Myofibers are cylinder shaped reaching 40 cm in length and 2500 µm in width and therefore are the cells in the human body with the larger volume. To be able to contract such a large volume synchronously, on a different scale than most human cells, myofibers have specialized structures called triads, the first described membrane contact site, composed of a plasma membrane invagination (T-tubule) in contact with the ER. While the importance of triad localization in proper muscle function is well established, the mechanisms underlying T-tubule and triad formation remain poorly understood. The Arp2/3 complex is a major branched actin nucleator. It consists of seven subunits, and in higher Eukaryotes, some subunits are encoded by more than one gene, giving rise to eight unique Arp2/3 complexes. This intriguing diversity raises questions about whether different Arp2/3 complexes possess unique properties tailored for a specific cellular function. We have previously shown that Arp2/3 plays a vital role in triad organization by an unknown mechanism. In this work, by performing Airyscan timelapse imaging in myofibers, we found that T-tubules are dynamic and that lack of Arp2/3 boosts T-tubule growth. The same boost in growth was observed when cortical actin was disrupted. At the molecular level, we observed that different Arp2/3 complex isoforms impact T-tubule growth differently, with the Arpc5 but not Arpc5L isoform depletion leading to increased T-tubule growth, T-tubule aggregates and reduced triad formation. At the physiological level, we observed that Arpc5 but not Arpc5L is important for synchronous calcium release and myofiber contraction. This work emphasizes the significance of the Arp2/3 complex and the Arpc5 isoform in regulating the intricate interplay between T-tubule organization, triad formation, and the proper execution of calcium release and contraction in cells with large volume.

Topic Modeling to analyze spatial transcriptomic data from Allen Human Brain Atlas

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Human brain is a complex interconnected structure controlling all elementary and high-level cognitive tasks. One of the main aims of neuroscience community in past decades has been to connect genetic information of anatomical structures to their underlying biological function. The availability of data set with vastly enhanced structural coverage allows an explicit approach aimed at identifying network structure common across individuals which is related to structural and functional organization of the entire brain. The Allen Human Brain Atlas (http://human.brain-map.org/) consists of microarray data in 3702 spatially distinct samples taken from six neurotypical adult brains. To understand large-scale transcriptome organization, we apply a Topic Modeling tool called hierarchical Stochastic Block Model (hSBM) to a set of highly variable genes. Topic models are a group of algorithms developed to infer the latent topical structure of a collection of documents. hSBM is a generative model and differs from other methods describing the problem as a community detection problem on a bipartite network, where one layer has samples as nodes, the other one has genes and links that connect them are weighted accordingly to the expression of each gene in each sample. The goal would then be to cluster similar brain samples and to identify sets of genes (topics) that characterize each ensemble, in order to compare samples organization into clusters with anatomical structure and functional connectivity. One of the key results of the work is that different levels of clustering present clusters composed quite homogeneously of samples coming from six donor brains. This is proof that the algorithm does not focus on inter-individual differences in gene expression but captures characteristics of the dataset that can be considered conserved between subjects. Furthermore, by comparing clustering results from all hierarchical levels with the organization in anatomically distributed brain regions, there are evidences of how the agreement differs strongly from that which would occur in a random case. At the end we developed a multipartite version of hSBM, that we call nSBM, designed to integrate gene expression data with other omics on the same samples, as well as methylation and mutation data or spatial information, that allows us to investigate more deeply and in a richer framework complex patterns in tissues such as brain ones.

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23 Exploring microbial community dynamics: Unravelling complex interactions in fluctuating habitats

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Microbes commonly thrive in large communities consisting of hundreds to thousands of species hinging on a complex network of interactions. The outcomes of microbial interactions shape the dynamics of life on Earth and significantly affect environmental and human health, as well as climate stability. The environment hosting microbes is typically subject to fluctuations in the concentration of chemical compounds and microbial communities are resilient from fluctuations ranging over a wide range of timescales. A comprehensive understanding of the mechanisms governing the dynamics underlying microbial communities' resilience is still lacking. While it is known that communities comprising two species experience variation in composition on timescales longer than the fluctuation timescales, we lack both experimental data and theoretical modeling regarding the spatiotemporal dynamics of complex bacterial communities, comprising more than two species, subject to fluctuations. This poster addresses a fundamental inquiry in ecology: How is complex microbial community dynamics affected by fluctuations in the chemical composition of their environment? I will show how to leverage microfluidic techniques to fluctuate nutrient concentrations within a micron-sized chamber carrying a three-species bacterial community. How I tracked cell growth rates and cell spatial position over time. First outcomes from experiments and numerical agent-based model (ABM) featured to explore the influence of the number of species and their relative spatial position on the community development will be presented. By spatially and temporally characterizing microbial interactions dynamics throughout fluctuating environments, this work brings about a more fundamental, mechanism-based understanding of microbial community resilience.

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24 NanoPyx: Accelerating bioimage analysis across different scales

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The field of bioimaging has witnessed groundbreaking advancements through automated analysis, offering unprecedented insights into life across diverse scales. From the microscopic scrutiny of single-cell microorganisms to the comprehensive examination of entire tissues, automated analysis ensures researchers have access to quantitative, unbiased, and reproducible methods. However, the analysis of extensive and intricate datasets has posed a significant challenge, hindering experimental efficiency and scalability. To overcome this challenge, we created NanoPyx, a Python-based framework aimed at overcoming the challenges associated with bioimage analysis. Central to NanoPyx is the Liquid Engine, an intelligent machine learning-based agent that significantly accelerates image analysis tasks. NanoPyx incorporates an extensive suite of super-resolution methods, including previously exclusive ImageJ plugins, covering drift correction, channel registration, SRRF/eSRRF, Fourier Ring Correlation, Image Decorrelation Analysis, NanoJ-SQUIRREL's error map for artifact detection, and more. NanoPyx's adaptive nature and self-optimization capabilities, exhibits significantly reduced runtimes compared to its predecessor, NanoJ. Additionally, NanoPyx caters to researchers of varying coding expertise, providing accessible access through Python libraries, interactive Codeless Jupyter Notebooks, and a seamless napari plugin. The self-optimization principles underlying the Liquid Engine hold promise for advancing high-performance computing in diverse research domains. NanoPyx emerges as a valuable tool empowering researchers to unlock the full potential of bioimaging data analysis, thereby fostering accelerated discoveries in biology from full tissues to subcellular components.

Using Single Molecule Tracking to investigate the regulation of Ascl1 by Post Translational Modifications during neurogenesis and neuronal reprogramming

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Vertebrate neurogenesis is to large extent regulated by proneural transcription factors (TFs) such as Ascl1, both required and sufficient to promote a full program of neuronal differentiation. In line with its master regulatory function in neurogenesis, Ascl1 is the most common player in TF cocktails used in neuronal reprogramming protocols. Post-translational modifications (PTMs) define a critical level of regulation of Ascl1 function. Multisite phosphorylation of Ascl1, downstream cell-cycle regulators, downregulates its activity in a proliferative context. Phosphorylation occurs at six serine-proline (SP) sites located outside the DNA-binding domain, in a rheostat-like model whereby the number - but not location - of phospho-residues determines Ascl1 activity, evoking a mechanism based on modulation of electrostatic potential. Among other things, electrostatic forces are often used by TFs while scanning the genome, thereby impacting sequence-specific binding to target genes. Protein O-GlcNAcylation (O-GlcNAc) is a metabolism regulated PTM, characterized by the addition of a (non-charged) O-GlcNAc moiety, to a Ser/Thr by an O-GlcNAc transferase (OGT). It is the current view that O-GlcNAc regulates protein function to large extent by counteracting phosphorylation at overlapping/neighboring

residues. This could provide a very effective mechanism of regulating Ascl1 function, a hypothesis we are currently addressing. Single Molecule Tracking (SMT) emerged as a state-of-the-art technique to survey complex interactions between TFs and chromatin, resulting in a highly quantitative understanding of TF binding dynamics in live cells. Here, we describe our ongoing efforts to understand how PTMs impact Ascl1 activity during development and neuronal reprogramming, at a single molecule resolution.

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26 **Generalized Lotka Volterra model on the Bethe lattice**

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Recent times have witnessed a burst of activity on the application of equilibrium and non-equilibrium statistical mechanics tools and ideas to study the behaviour of large ecosystems, in particular their stability and the nature of their equilibria. In particular, many results on the coexistence of many species have been obtained using the generalized Lotka-Volterra model. The latter, under appropriate hypothesis on the shape of the interaction matrix between species and the stochasticity concourring to the dynamics (demographic noise), allows to recast the dynamical stability problem in terms of equilibrium statistical mechanics. We present here for the first time results on the equilibrium statistical mechanics of the generalized Lotka-Volterra model with an interaction network between species which is sparse (Behe lattice). Our analysis, at variance with the standard approach which makes use of a less realistic dense intra-species interaction network, reveals novel and highly non-trivial heterogeneity effects in the populations distributions, as for instance strong deviations from Gaussianity when increasing the heterogeneity of intra-species interactions. In this talk I will review the method exploited to study the problem, i.e., how the effective Hamiltonian for species interactions derived in [A. Altieri, F. Roy, C. Cammarota, and G. Biroli, Phys. Rev. Lett. 126, 258301 (2021)] can be used to generate local marginals for populations distribution abundance when interactions are sparse, and I will present the main results obtained varying both the temperature (strength of demographic noise) and the heterogeneity of interactions

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The cost and evolutionary advantages of aneuploidy in saccharomyces cerevisiae 27

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Aneuploidy refers to an abnormal number of chromosomes in a cell. Aneuploidies have been associated with cellular and organismal defects. Lately, it has been proposed that aneuploidy may confer selective advantages to cells under specific conditions. A fine balance between the benefits and costs of aneuploidy has been proposed to explain these evolutionary dynamics. However, the general principles governing the costs of aneuploidy, and the molecular and cellular processes at their basis are still obscure. We aim to precisely characterize the costs of aneuploidy in S. cerevisiae by combining theoretical, analytical, and experimental approaches.

SR34a: a plant splicing factor crossing the nuclear envelope to regulate mRNA translation? 28

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Serine/arginine-rich (SR) proteins are members of a highly conserved family of RNA-binding proteins that regulate alternative splicing and, in plants, modulate responses to stress. A few Arabidopsis thaliana SR proteins exhibit nucleocytoplasmic shuttling activity, indicating that they may fulfil post-splicing roles, such as mRNA export, stability or translation, as has been shown in animal systems. We are investigating whether SR34a, an arabidopsis nucleocytoplasmic shuttling SR protein, plays a direct role in mRNA translation. Using polysome fractionation through sucrose gradients, we found that SR34a associates with actively translating ribosomes. We also generated transgenic arabidopsis lines expressing a functional nuclear-retained version of SR34a in which the SR protein's shuttling activity is lost but its splicing function is maintained. Importantly, these plant lines phenocopy an sr34a loss-of-function mutant, indicating that export from the nucleus is required for the SR protein's role in stress tolerance. We are currently performing RNA-sequencing to compare the translatome (mRNAs isolated from polysome fractions) of wild-type plants with that of the SR34a nuclear-retained lines to establish whether the SR protein functions in translational regulation. Our work is expected to identify the first plant SR splicing factor playing a broader role in gene expression regulation.

Characterisation of swimming phenotypes using parallel microscopy

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IGC

Chlamydomanas reinhartdii (CR) cells exhibit a fascinating ability to navigate towards light. Other important phenotypic traits like growth and cell division number are also strongly correlated with light. We aim to explore the diversity and dynamics of phenotypes expressed by CR cells by systematically perturbing the genetic background and light environment of these cells.

To achieve this, we embark on a microscopy marathon that will encompass a total of ≈900 days of microscopy time. Here, we present the experimental methodology behind our study.
