Book of Abstracts

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A SYNTHETIC EPIGENETIC CIRCUIT PRODUCES NOISE-DRIVEN HERITABLE OSCILLATIONS

Amith Zafal Abdulla
Brandeis University, USA

ABSTRACT
In eukaryotes, heritable information is not only genetically encoded but is also stored in the form of covalent histone modifications that can activate or repress transcription. We do not understand how different sources of noise impact the transmission of epigenetic information. Here, we engineer a synthetic negative feedback circuit in fission yeast in which a repressive histone-modifying enzyme suppresses its own expression. Mathematical modeling predicts a stable fixed point with spiraling dynamics, yet in the regime of histone-modification fluctuations, we observe the emergence of sustained noise-driven oscillations. Experimentally, approximately 5% of cell lineages exhibit heritable oscillations persisting over 40 generations, while the remainder stochastically switch between high and low expression states. We show that positive feedback associated with repressive modifications suppress these fluctuations, directly linking epigenetic inheritance to buffering noise. Remarkably, oscillatory dynamics propagate to distal genes, producing correlated oscillations across genomic loci. Our results reveal how noise can produce sustained, heritable oscillations in an epigenetic circuit where spreading of histone modifications drive correlated expression patterns across spatially distant genes.

KEYWORDS
Epigenetics, Synthetic circuit, Negative feedback

HFSP Reference Number: LT0020/2024-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2024

RNA-INTERACTOME CAPTURE ISOLATES A BACTERIAL EFFECTOR THAT MODIFIES HOST MESSENGER RNA TO PROMOTE VIRULENCE

Frederic Allain
Swiss Federal Institute of Technology in Zurich (ETH Zürich), Switzerland

Stefan Gaussmann[1]; Kathleen Mccaffrey[2]; Garrett Ng[2]; Tristan Dewald[3]; Mark Helm[3]; Elizabeth Hartland[2]
[1] Swiss Federal Institute of Technology in Zurich (ETH Zürich); [2] Monash University; [3] Johannes Gutenberg-University Mainz

ABSTRACT
Bacterial pathogens utilize specialized secretion systems to translocate effector proteins into host cells, where they manipulate cellular processes to promote infection. Although many bacterial effectors post-translationally modify host proteins, whether they directly target host RNA remains largely unexplored.
Here, we used RNA-interactome capture (RIC) during Legionella pneumophila infection of human macrophages to identify bacterial effectors associated with host messenger RNA (mRNA). This approach revealed a previously uncharacterized effector, FadA (Lpg1666), which localizes to the host cell nucleus and interacts with newly synthesized host mRNAs. We determined the three-dimensional structure of FadA by X-ray crystallography. The structure shows that FadA adopts a canonical Rossmann fold that binds a single flavin adenine dinucleotide (FAD) molecule within a large, positively charged pocket. Biochemical analysis demonstrated that FadA functions as a highly efficient oxidase that generates reactive oxygen species (ROS) in a NADPH-dependent manner. This oxidase activity promotes the formation of 8-oxoguanine (8-oxoG) modifications on associated mRNA substrates. Cross-linking immunoprecipitation sequencing (CLIP-seq) revealed that FadA binds host mRNAs at uracil-rich VUUUK sequence motifs. The resulting 8-oxoG modifications damage host mRNAs and reduce their translation at the ribosome by approximately 80%. Among the affected transcripts are mRNAs encoding inflammatory transcription factors, including NF-κB and AP-1. By suppressing cytokine responses, FadA promotes L. pneumophila replication within macrophages and supports bacterial persistence in vivo. Together, these findings uncover a previously unrecognized mechanism of bacterial virulence in which a secreted effector post-transcriptionally modifies host mRNA to suppress immune responses

KEYWORDS
Legionella pneumophila, Bacterial effector proteins, Host mRNA modification, 8-oxoguanine, Immune evasion

HFSP Reference Number: RGP013/2023
HFSP Award Category: Research Grant
HFSP Award Year: 2023

UNRAVELLING THE ROLE OF PROBOSCIS MECHANOSENSORY NEURONS IN AEDES AEGYPTI BLOOD FEEDING BEHAVIOR

Irene Arnoldi
Radboud University Medical Center, Neetherlands

Felix Hol
Radboud University Medical Center

ABSTRACT
The yellow fever mosquito Aedes aegypti performs blood feeding on human hosts to complete egg development, potentially transmitting pathogens at the site of bite, through the injection of infected saliva. After landing on the skin of the host, female mosquitoes start probing this tissue for a blood vessel to pierce, exploiting a plethora of different thermal, mechanical and chemical stimuli. Cues stimulate sensory neurons located in the labrum, the only stylet harboring cells of the six composing the fascicle which penetrates the skin during biting. The labrum carries four pairs of sensilla, insect specialized sensory organs, and constitutes the food channel through which the blood flows during engorging. Within this sensory arsenal, two pairs of sensilla, the labral ridge receptors and the campaniform sensilla, have been previously suggested to have a mechanosensory function. To unveil which cells are involved in the processing of mechanosensory cues during biting, we selected different gene markers potentially specific for the mechanosensory neuronal populations of the labrum and we checked their expression during adult development by in situ hybridization and quantitative polymerase chain reaction. Concurrently, we are developing an assay to check the activation of labral neurons after providing a mechanosensory stimulus and using a genetically modified mosquito strain expressing the calcium sensor GCaMP7s under the control of the pan-neuronal marker brunchpilot.

KEYWORDS
Mosquito, mouthparts, mechanosensory processing, neurons

HFSP Reference Number: LT0040/2025-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2025

HARMONY IN DYNAMICS: TRANSLATING A PARTICIPATORY ARTWORK INTO A SOCIO-COGNITIVE EXPERIMENTAL PARADIGM

Hasan Ayaz
Drexel University, USA

Uri Hershberg[1]; Goni Shifron[2]; Luciano Perondi[3]
[1] University of Haifa; [2] Multi-disciplinary independent artist; [3]Iuav University of Venice

ABSTRACT
Collaborations between art and science often remain illustrative or interpretive, with one discipline serving the other. In contrast, true art-science collaboration requires co-development of a shared framework in which artistic complexity is preserved while scientific rigor is introduced. Here, we describe the transformation of the participatory artwork Tä, created by artist Goni Shifron, into a socio-cognitive experimental paradigm for studying the emergence of group dynamics through joint interaction with a shared environment. In Tä, participants sit around a table with several piles of pebbles and follow simple rules for manipulating them. Without speaking or directly communicating, individuals initially engage in independent actions, but over time their behaviors become increasingly coordinated and collective patterns of movement emerge. This setting provides a unique platform for investigating how synchronization and shared action arise in groups under minimally structured conditions. Our collaboration uses Tä as the core experimental environment to address two central questions: (1) how behavioral synchronization emerges during participation in Tä, and (2) whether the emergence of shared action reflects only moment-to-moment interpersonal adaptation or a form of group learning that transfers to subsequent cooperative interaction. To examine these questions, each session will include 6 to 12 active participants, a comparable number of audience members, a trained host serving as facilitator, and observers. Participants and audience members complete questionnaires before and after the Tä experience and engage, in separate groups, in an additional cooperative task designed by Shifron. This design allows comparison of the effects of direct participation versus observation on subsequent group coordination. By embedding experimental measures around an intact participatory artwork, this project establishes a novel framework for studying collective behavior, synchronization, and transfer of group learning without reducing the aesthetic and social richness of the original piece. More broadly, it offers a model for rigorous art-science collaboration in the study of emergent human dynamics.

KEYWORDS
Multidisciplinary research, Art and Science, self organization

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HFSP Award Category: Research Grant
HFSP Award Year: 2025

A TALE OF TAILS: INVESTIGATING THE CROSS-COMPATIBILITY AND ANCESTRY OF THE BACTERIAL FLAGELLAR FILAMENT.

Matthew AB Baker
UNSW Sydney, Australia

Jacob Scadden[1]; Divyangi Pandit[1]; Pietro Ridone[1]; Yoshiyuki Sowa[2]
[1]UNSW Sydney; [2]Hosei University

ABSTRACT
Motility and chemotaxis are major adaptations that have allowed microbial life to expand to nearly every environment on Earth. One of the major mechanisms of bacterial motility utilises the bacterial flagellar motor (BFM) to drive movement. Understanding the evolution of the flagellar filament protein, flagellin, is important in elucidating the diversity observed within this protein and its adaption in various environments to provide motility. We calculated phylogenetic trees using sets of representative flagellin sequences to estimate ancestral sequence reconstructions (ASR) of ancestral flagellin proteins. Candidates were cloned into Escherichia coli K-12 and Salmonella enterica serovar Typhimurium strains which had the flagellin gene(s) deleted. All ASR candidates did not rescue motility in E. coli, however, motility was rescued in S. Typhimurium motility by specific ancestral flagellins. To test the compatibility and interchangeability of flagellin domains from different species, we designed flagellin chimeras and expressed these in filament-deleted E. coli (ΔfliC). Two of the eleven outer domain chimeras, as well as outer domain deleted flagellin variants, were able to form filaments and were motile. Overall we show that the outer domains are not necessary for motility in E. coli K-12 and that rational engineering of outer domains help to identify key residues that are maintained in flagellar filaments.

KEYWORDS
bacterial motility, flagellin, ancestral reconstruction, complex systems.

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HFSP Award Year: 2021

MAPPING STRUCTURAL AND FUNCTIONAL CONNECTIVITY OF THE DISTRIBUTED SENSORY SYSTEM IN CHITON ARMOR

Daniel Baum
Zuse Institute Berlin, Germany

Ling Li[1]; Daniel Baum[2]; Daniel Speiser[3]; Chenhao Hu[1]; Adriana Halvonik-Sanchez[3]; Tanvi Deshmukh[1]; Kira Schmitt[2]; Yanbin Li[1]
[1]University of Pennsylvania; [2]Zuse Institute Berlin; [3]University of South Carolina

ABSTRACT
Many mollusks produce hard, mineralized shells for mechanical protection. Chitons are unique among extant mollusks in that their biomineralized shell plates incorporate living tissue. These innervated tissues, known as aesthetes, form complex three-dimensional networks that fill thousands of long, narrow channels running through the shell plates and extending to the shell surface. Aesthetes serve several sensory functions, particularly photoreception. Across chiton species, aesthetes include visual elements with varying levels of complexity, ranging from simple photoreceptors to eyespots to image-forming eyes with mineralized lenses that provide spatial vision. Together, these elements constitute a distributed sensory network (DSN) embedded within the mineralized shell plates. This "hard-wired" sensory architecture combined with decentralized visual processing contrasts with most visual systems in biology, which rely on a small number of specialized sensory organs and centralized neural processing. While previous studies have focused on overall behavioral responses of animals and the fine structure of individual sensory elements, the system-level organization and function of chiton DSNs remain largely unknown. Our HFSP project aims to understand how these networks are wired, how they function as integrated systems, and how resilient they are to damage. Here we present the progress of ongoing research, particularly focusing on developing a quantitative digital representation of the chiton DSN at the entire-shell level. Using high-resolution X-ray tomography together with advanced 3D data segmentation, registration, and analysis, we reconstructed the complete DSN network of Chiton marmoratus, a species of chiton with eyespots. The reconstruction reveals a distinct arrayed, tree-like network architecture. Specifically, the anterior valve contains over 14,000 eyespots (~450 eyespots/mm2) arranged in a pseudo-hexagonal pattern on the shell surface. These eyespots connect into local clusters that merge into larger "trunks", which organize into radial rows and ultimately link to the animal's decentralized nervous system. We further represent the DSN as a physical graph network (total length ~800 mm) and analyze its wiring pattern and efficiency using graph-theoretic approaches such as Steiner network analysis. Our preliminary results suggest that the chiton DSN does not simply minimize total wiring length. Instead, its architecture likely reflects a balance between sensory performance, mechanical protection, developmental constraints, and evolutionary constraints.

KEYWORDS
Biological materials, marine invertebrates, visual system, imaging, animal behavior, computer vision

HFSP Reference Number: RGP016/2023
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HFSP Award Year: 2023

CHROMATOPHORES UNCOVERED FROM GENES TO CELLS

Pierre Beaudier
University of Geneva, Switzerland

Adrian Lutz[2]; Pierre-Yves Helleboid[1]; Devi Stuart-Fox[2]; Athanasia Tzika[1]
[1]University of Geneva; [2]University of Melbourne

ABSTRACT
Animal colouration is remarkable in the breadth of functions it conveys, from camouflage to display and from thermoregulation to social communication. Numerous studies focus on mammalian coloration, which solely relies on melanin-producing cells in the skin. Yet, the strikingly colorful skin of reptiles has received comparatively less attention. We used a multidisciplinary approach to broaden our understanding of such phenotypic diversity by investigating two color morphs of the corn snake, a species characterized by bright red and orange skin coloration. Using genetic mapping, we identified the mutations responsible for the yellow and grey coloration of the Caramel morph and the predominantly vibrant red coloration of the Sunkissed morph. These findings were confirmed by functional analyses using our gene-editing protocol in corn snakes. We coupled our results with electron microscopy imaging to investigate subcellular modifications of the color-producing cells, as well as extensive pigment analyses to determine their composition. We focused in particular on xanthophores, which convey red and yellow hues through their pigment content. Although much is known about the maturation and subcellular organization of melanophores, and an expanding body of literature addresses iridophores, which contain reflective guanine crystals, knowledge of xanthophores remains limited. We sought to fill this gap by relating the modified hue observed in the Caramel and Sunkissed morphs, compared to the wild type, to changes in their transcriptomic profiles. Furthermore, we established expansion microscopy imaging protocols that will enable us to elucidate the content of the pigmented organelles, which remains poorly understood. Together, our findings provide new insight into the genetic and cellular mechanisms generating color diversity in reptiles and establish a framework for future studies of chromatophore biology.

KEYWORDS
Chromatophores, skin coloration, pigment cells, phenotypic diversity

HFSP Reference Number: RGP0037/2022
HFSP Award Category: Research Grant
HFSP Award Year: 2022

REAL-TIME SINGLE-MOLECULE IMAGING IN ZEBRAFISH EMBRYOS UNCOVERS NON-CANONICAL TRANSLATION

Maelle Bellec
Max Planck Institute for Heart and Lung Research, Germany

ABSTRACT
Spatiotemporal control of protein synthesis is essential for many developmental processes, yet determining when and where specific proteins are produced in vivo remains a major challenge, particularly in vertebrate embryos. The translation of bmp2b, a key dorsoventral morphogen, is known to be tightly regulated during early development, but the translational step underlying this regulation has remained unclear. Using ALFA_array labeling combined with lattice light-sheet microscopy, we visualized bmp2b translation in real time at single-molecule resolution in living zebrafish embryos. We
extended this platform to simultaneously track individual mRNAs and their translation in live embryos using dual-color imaging. This enables the precise spatial mapping of translating mRNAs in vivo and provides a critical validation of translation sites observed in our assays. Quantitative analysis revealed that bmp2b translation is primarily limited at the level of ribosome initiation rather than elongation. Consistently, substitution of native bmp2b UTRs with viral sequences strongly increased both the fraction of translating mRNAs and the translation efficiency per transcript. Additional experiments using viral and actb2 5′UTRs suggest the involvement of a cap-independent initiation mechanism. Together, this developed system provides direct insights into translational regulation in vivo and highlights the complexity of post-transcriptional control in early vertebrate development.

KEYWORDS
Live imaging, translation, embryo, single-molecule

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THE BLUE-WINGED EYES OF MORPHO BUTTERFLIES

Gregor Belusic
University of Ljubljana, Slovenia

Uroš Cerkvenik[1]; Marko Ilić[1]; Aleš Škorjanc[1]; Jack Supple[2]; Andrew Dang[3]; Sofia Diaz[3]; Adriana Briscoe[3]; Primož Pirih[4]
[1]University of Ljubljana; [2]Imperial College London; [3]University of California, Irvine; [4]Salzburg University

ABSTRACT
Morphos are very large butterflies that inhabit the endless rain forests of Central and South America. Most species are famous for their blue wing coloration and vividly react to blue objects. However, their wing color varies widely (from blue to brown and white) among sympatric species with distinct circadian activity, inhabiting different forest strata. This raises questions of whether and how their visual systems facilitate species-specific communication across ecological niches. To investigate how ecological factors shape signal perception, we studied nine Morpho species coexisting in French Guiana that differ in flight height (canopy vs. understory) and circadian activity. Using a custom insect ophthalmoscope, intracellular and extracellular electrophysiology, immunohistochemistry and anatomical analyses, we examined their retinal specializations. All studied butterflies have very large compound eyes which have a high sensitivity and acuity, allowing for long distance detection of mates and competitors. The UV-, blue-, green- and red-sensitive photoreceptors support multispectral color vision and form ordered retinal mosaics with at least six types of optical units, the ommatidia. Similar mosaics are found across many other nymphalid subfamilies and are based upon three visual pigments - UV, blue (B) and long-wavelength (LW) opsins. However, Morphos have triplicated LW opsins, two of which are either blue- or red-shifted. The blue-shifted LW opsin and a blue opsin together form two receptor classes that support a dense sampling of the blue-green spectrum. The red-shifted LW opsin, together with filtering pigments, is the basis for photoreceptors with low short-wavelength sensitivity that create neural signals decorrelated from signals from blue receptors, allowing the brains to efficiently compute colors. Furthermore, some species have UV and blue photoreceptors with very high polarization sensitivity. These feed into high-order neurons that function as target detectors, sensitive to blue moving objects and reacting differently to polarized and colored stimuli. While built on a generalist scheme, typical for all butterflies from the family Nymphalidae, the eyes of Morphos contain numerous small evolutionary optimizations for viewing conspecifics.

KEYWORDS
Morpho, compound eyes, color vision, opsin, polarization vision

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LIFE IN THE MAZE: MICROCLIMATE VARIABILITY CONTRIBUTES TO THE LOCAL COEXISTENCE OF FYNBOS ANTS

Tom Rhys Bishop
Cardiff University, UK

Phillip Hoenle[1]; Brittany Trew[2]; Rebecca Senior[3]; Andrew Davies[2]; Charlene Janion-Scheepers[4]
[1]Cardiff University; [2]Harvard University; [3]Durham University; [4]University of Cape Town

ABSTRACT
Imagine your favourite location in nature. It could be a forest, a grassland, or shoreline. Regardless of where your imagination takes you, we can be sure of one thing: there will be many different species coexisting there. You will find a range of birds in the forest, many different spiders in the grassland, and a diversity of crabs within the rocky shore. Across nearly all landscapes of Planet Earth, species coexist. Why is this?
Traditionally, scientists have tried to explain species coexistence by focussing on the differences that species have. For instance, different species eat different things, or they may nest in slightly different parts of the environment. These differences prevent species from directly competing with each other for resources, and this lack of competition means that they can coexist peacefully. This way of thinking has dominated the way that scientists view species coexistence. Species differences, however, cannot always explain coexistence patterns. Repeatedly, we find examples where many species coexist despite their overlapping food, nesting or foraging requirements. How can this be? Here, we propose a simple, new idea for these difficult-to-explain coexistence cases. Most organisms are small and, from their perspective, their environments are dynamic “mazes” of micro-environments. These mazes of micro-environments vary in four dimensions: left, right, up, down, and through time. Crucially, species may become isolated from each other in their own corner of these mazes. As this happens, the chances of competition decrease, and the chances of coexistence across the landscape as a whole increase. We seek to test this "micromaze" hypothesis. To do this, we focus on the diverse ant (Hymenoptera: Formicidae) communities of the fynbos biome in South Africa. First, we use a range of field and lab techniques to estimate the realised and potential temperatures under which >30 ant species can forage. Second, we pioneer a novel mechanistic microclimate modelling method that combines drone-based LiDAR data on the vegetation and topographic structure of our field sites, with empirical sensor data, to estimate the hourly near-ground microclimate at 2 m x 2m gridded resolution.
Our initial results reveal that the foraging activity of ants is driven by a combination of macro and microclimatic factors, but that greater spatiotemporal microclimate diversity leads to greater numbers of ant species coexisting on our 0.25 hectare study plots. Consequently, we provide some of the first evidence that microclimatic variability contributes to the coexistence of species at a fine spatial scale. Moving forward, we seek to link spatially explicit data on individual ant colonies with their estimated foraging distances to further test the biological mechanisms that underlie the micromaze hypothesis.

KEYWORDS
coexistence, ecology, biodiversity, microclimate, temperature

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MECHANISM OF SPLICEOSOME TERMINATION

Vytaute Boreikaite
Research Institute of Molecular Pathology (IMP), Austria

Matthias Vorländer[1]; Alexander Phillips[1]; Rupert Faraway[1]; Leonie Opitz[1]; Moritz Wanke[2]; Gerald Raffl[2]; Laura Fin[1]; Stefan Ameres[2]; Clemens Plaschka[1]
[1]Research Institute of Molecular Pathology (IMP); [2]University of Vienna

ABSTRACT
Splicing of precursor messenger RNA (pre-mRNA) is essential for eukaryotic gene expression and involves excision of introns and ligation of exons to produce a mature protein-coding mRNA. Pre-mRNA splicing is catalysed by a large and dynamic RNA-protein machine called the spliceosome. After intron excision, the spliceosome becomes trapped in a non-productive complex bound to the branched RNA "lasso" known as the intron-lariat. This spent spliceosome complex must be disassembled to liberate the spliceosome for subsequent catalytic cycles. Simultaneously, the intron-lariat must be extracted from the spliceosome's active site, debranched, or linearised, and finally released from the spliceosome to enable intron RNA decay. Together these events are referred to as spliceosome termination. Despite its importance for maintaining splicing efficiency and ensuring intron turnover, the underlying mechanism of spliceosome termination has remained elusive. Here, I will present our recent cryo-electron microscopy structures of native human spliceosomes captured at two sequential stages of spliceosome termination, which we find to be orchestrated by three ATP-driven RNA helicases and seven non-enzymatic protein factors. We show that two RNA helicases – DHX15 and Aquarius – begin the unwinding of the spliceosome's RNA active site, extracting the buried intron-lariat branch junction from the catalytic core of the spliceosome. This extraction event makes the branch junction accessible to the spliceosome-tethered debranching enzyme, which linearises the intron-lariat on the spliceosome. These remodelling events generate a novel spliceosome intermediate, which we name the debranched intron spliceosome (DIS). In turn, DIS recruits the third RNA helicase, DHX35, which ejects the now-linear intron from the spliceosome, driving spliceosome disassembly and enabling intron turnover. Notably, many of the same proteins – including the RNA helicases DHX15 and DHX35 – are also involved in the termination of defective spliceosomes stalled on faulty introns during spliceosome quality control. These similarities suggest a conserved mechanism for terminating the spliceosome after both successful and failed splicing events. Taken together, our structures of the native human spliceosomes, supported by biochemical, genetic and RNA-sequencing data, reveal the mechanism of spliceosome termination, which sustains accurate and efficient pre-mRNA splicing and, therefore, gene expression.

KEYWORDS
Gene expression, RNA, splicing, cryo-EM

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RECORDING THE HISTORY OF CELL-CELL INTERACTIONS IN VIVO

Costanza Borrelli
The Rockefeller University, USA

ABSTRACT
Cellular interactions underlie most biological processes in multicellular organisms. To dissect how cells communicate with each other in homeostasis and disease, our lab has established LIPSTIC (universal Labeling Immune Partnerships by SorTagging Intercellular Contacts), a method based on proximity-dependent enzymatic labeling across cell-cell membranes (1, 2). LIPSTIC involves expression on donor cells of the Staphylococcus aureus transpeptidase Sortase A (SrtA), which covalently transfers a labeled substrate containing the sorting domain LPETG to five N-terminal glycine residues (G5-tag) on acceptor cells. uLIPSTIC transgenic mice allow for in vivo detection of the interactors of any cell type that can be turned into a LPTEG-tag donor by specific Cre-recombination, and has been used by us and others to dissect interactions between several immune and non-immune cell types (1–5). Due to the transient nature of its labelling, however, LIPSTIC experiments provide information about interactions occurring at the time of substrate administration, and are therefore a snapshot representation of a cell's interactome. To experimentally link cell-cell interaction events to downstream cellular behaviors and fate decisions, we will leverage the versatility of lipid nanoparticles (LNPs) to deliver mRNA encoding recombinases or gene editing enzymes to LIPSTIC labelled cells, using the transferred LIPSTIC substrate on the surface of interacting cells as a docking site for LNPs. The LIPSTIC-labelled interaction will therefore be permanently encoded in the targeted cell's genome, for example by recombinase-mediated STOP cassette excision and consequent expression of the fluorescent reporter. This permanent mark can subsequently be read out by flow cytometry long after the interaction has occurred and the transient LIPSTIC labelling on the cell surface has faded. To engineer a system in which up to 10 cellular interaction events can be sequentially recorded into the genome of interacting cells, we will then combine the LIPSTIC technology with in vivo DNA recording in the DARLIN mouse (6). To record cell-cell interaction events, rather than lineage, in the cell genome, we will take advantage of one of the DARLIN target arrays and use it as a cellular memory device. Instead of simultaneously expressing all 10 sgRNAs, we will sequentially deliver sgRNA 1-to-10 to LIPSTIC labelled cells, inducing time-controlled and interaction-dependent scarring at each of their target sites. As each scarring event will result in a distinct repair pattern which will be inherited by the progeny of interacting cells, a lineage history of the interacting cells can also be derived. Reconstructing the history of cell-cell interactions will allow us to link cellular communication events to cell fate decisions, deepening our knowledge of the molecular rules that govern the biology of complex organisms.

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KEYWORDS
cellular interactions, LIPSTIC, LNPs

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NEW KIDS ON THE BLOCK: HOW DENOVO EMERGED MICROPEPTIDES REWIRE CELLULAR NETWORKS

Anne-Ruxandra Carvunis
ETH Zurich, Switzerland

Christine Brun[2]; Erich Bornberg-Bauer[3]; Jiwon Lee[4]; Aaron Wacholder[1]; Anna Grandchamp[2]; Mathilde Slivak[2]; Hugo Bonnefous[2]; Margaux Aubel[3]; Sarah Lucas[3]
[1] ETH Zurich; [2]University of Aix-Marseille; [3]University of Muenster; [4]University of Pittsburgh

ABSTRACT
Our understanding of protein evolution has recently been reshaped by the realization that entirely new genes can evolve de novo from previously non-coding DNA, rather than by modification of ancestral genes. The proteins encoded by these genes represent true molecular “new kids on the block”, with sequences and structures that have no evolutionary precedent. De novo proteins arise so frequently that thousands have been identified in the genomes of species spanning the tree of life. While most are rapidly lost, many de novo proteins persist and acquire biological relevance, some even becoming essential. This raises a fundamental but largely unexplored question: how do certain newcomers, but not others, coexist with the densely interconnected and evolutionarily ancient cellular “block” they enter? Proteins act within complex interaction landscapes, yet de novo proteins are usually studied in isolation. Here, we investigate how recently emerged human de novo proteins integrate and potentially reshape pre-existing molecular interaction networks. We first map the coding landscape of the human genome beyond known genes by integrating large-scale transcriptomics and translatomics data. We then apply a stringent comparative genomics pipeline to define a high-confidence set of human de novo proteins and reconstruct their evolutionary histories. Finally, we explore features that may influence cellular compatibility by combining computational predictions of protein interactions with analysis of predicted biophysical properties and population genetic variations. Our work offers an emerging view of how cellular systems tolerate continual molecular innovation, and how the established "block" constrains or enables the success of the "new kids".

KEYWORDS
protein evolution, molecular networks, translation, peptides, protein-protein interactions, primates

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HFSP Award Category: Research Grant Program
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INVESTIGATING CD1A-DRIVEN T CELL RESPONSES TOWARD UV OXIDISED SKIN LIPIDS IN HEALTH AND DISEASE.

Laura Ciacchi
University of Oxford, UK

Clare Hardman[1]; Karmella Naidoo[1]; Julian Moody[2]; Jia Jia Lim[3]; Adam Shahine[3]; Tan-Yun Cheng[4]; Yi-Ling Chen[1]; Branch Moody[4]; Jamie Rossjohn[3]; Fmedsci Graham Ogg[1]
[1]University of Oxford; [2]Tufts University; [3]Monash Biomedicine Discovery Institute; [4]Harvard Medical School

ABSTRACT
Ultraviolet (UV) radiation can drive skin inflammation in specific clinical photosensitivity settings such as polymorphic light eruption (PLE) and photoallergic dermatitis. CD1a is a monomorphic HLA class I-like molecule which is expressed at constitutively high levels by skin Langerhans cells (LC) and presents lipid antigens to T cells. Evidence is increasing for a role for CD1a in inflammatory skin disease pathogenesis through presentation of agonist and antagonist lipid antigens (Huang, et al. Cell 2023; Chen et al. Sci Immunol 2023). The effects of UV on skin lipids and the role of CD1a in health and disease are largely unknown. The overall aim of this study is to discover lipid antigens and uncover mechanisms that drive unconventional skin lipid-reactive T cells in health and disease. Through understanding aberrant self-reactivity towards oxidised skin lipids holds therapeutic promise to counteract the effect through antioxidants and replenishing depleted lipids at the skin barrier. Overall, by defining UV-altered neo-lipids and responsive T-cells will allow us to uncover associated mechanisms underlying UV-induced skin inflammation.

KEYWORDS
Skin, T cell immunology, UV radiation, photosensitivity

HFSP Reference Number: LT0028/2025
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2025

A DISTRIBUTED PRESSURE-SENSING PLATFORM FOR LONG-DURATION MAPPING OF FISH-GENERATED HYDRODYNAMIC SIGNATURES

Porcon Corentin
École Normal Superieur Rennes (ENS Rennes), France

James Herbert-Read[2]; Bastien Lagneaux[1]; Gurvan Jodin[1]; Florence Razan[1]
[1] ENS Rennes; [2] University of Cambridge

ABSTRACT
Understanding how aquatic animals perceive and respond to hydrodynamic cues requires experimental tools capable of resolving low-amplitude pressure fluctuations over biologically relevant spatial and temporal scales. Recent advances in embedded electronics have enabled the development of custom pressure-sensing instruments capable of capturing the subtle hydrodynamic traces left in the wake of swimming fish, helping to clarify how such signals may contribute to social interactions. We present a modular distributed pressure-sensing network designed to characterize fish-generated hydrodynamic signatures in confined aquatic environments. The system consists of 96 differential pressure sensors distributed over a 1 m² wall-mounted array, synchronized through a wireless time-coherent acquisition architecture operating at 200 Hz. The platform achieves a noise floor of ±0.15 Pa, a dynamic range of ±500 Pa, and an effective bandwidth of 0.3–30 Hz, enabling the detection of weak, spatially distributed pressure perturbations generated by freely swimming fish. Unlike conventional point-wise hydrophones or optical velocimetry techniques, the proposed approach enables long-duration, spatially resolved mapping of pressure fields with minimal intrusion and manageable data volumes. A dedicated synchronization framework integrates pressure recordings with high-speed video tracking, allowing reconstruction of spatio-temporal pressure footprints associated with individual trajectories. Long-duration experiments demonstrate the ability to capture evolving hydrodynamic wakes and transient pressure structures generated by swimming fish. This platform provides a new experimental basis for investigating lateral-line-mediated perception and hydrodynamic interactions in aquatic systems.
The modular architecture is scalable up to 240 sensing points and adaptable to different tank geometries, offering a versatile tool for interdisciplinary research at the interface of fluid dynamics, sensory biology, and behavioral ecology.

KEYWORDS
Hydrodynamic sensing, Fish locomotion, Pressure field mapping, Fish behavior, Pressure measurements, Fluid–structure interactions

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HFSP Award Category: Research Grant Program
HFSP Award Year: 2022

DRIVERS FOR TRANSPORT IN THE GASTROASCULAR NETWORK OF THE JELLYFISH AURELIA AURITA

Annemiek JM Cornelissen
CNRS & Paris Cité University, France

Laureline Julien[1]; Xinbo Li[2]; Aaron Winn[2]; Daisuke Noto[2]; Thibault Chasserau Annemiek[1]; Léna Zig Annemiek[1]; Mariam Ouali[1]; Alain Ponton[1]; Bérengère Abou Annemiek[1]; Eleni Katifori[2]
[1]CNRS & Paris Cité University; [2]University of Pennsylvania

ABSTRACT
The jellyfish Aurelia aurita possesses a gastrovascular system that digests and distributes nutrients. Its organs are arranged in the subumbrella as a quasi–two-dimensional disc containing four central stomachs connected to a canal network. From the stomachs, eight radial canals transport food toward a ring canal at the umbrella margin, from which a branched network redistributes nutrients back toward the center. We investigate the mechanisms driving transport within this system.
We identify two candidate drivers: (1) peristaltic deformations of the canals generated by the pulsatile muscle contractions used for swimming, and (2) beating cilia lining the canal walls. Electron and video microscopy show that each epithelial cell carries a single motile cilium coordinated in metachronal waves. Microrheology measurements reveal that the canal fluid is biphasic, consisting of seawater and mucus. Rheological characterization of extracted mucus demonstrates strong shear-thinning behavior, facilitating transport by lowering resistance under ciliary forcing. To separate the contributions of cilia and peristalsis, we track food particles in canals of jellyfish placed ex-umbrella down. Under menthol anesthesia, contractions cease and flow is driven solely by cilia. Comparing velocities in anesthetized animals with those in mildly contracting, recovering jellyfish shows that cilia maintain a baseline flow, while peristaltic contractions significantly accelerate transport. During resting phases between contractions, velocities return to anesthetized levels. To quantify how swimming-induced deformations drive flow, we develop a mathematical model of “length-induced” peristalsis that maps whole-body deformation to fluid propulsion. The model predicts that symmetry breaking is required to generate net transport and shows that different swimming modes can enhance forward flow or reverse it, enabling redistribution or expulsion of material. We are currently incorporating measured 3D body deformations of freely swimming jellyfish into the model to obtain quantitative predictions. Together, these results reveal a mechanism by which swimming kinematics control both the magnitude and direction of flow in the canal network. Ongoing work examines how mucus rheology and ciliary activity interact across developmental stages to regulate transport.

KEYWORDS
flow transport networks, jellyfish, cilia, peristaltis, mucus

HFSP Reference Number: RGP015/2023
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HFSP Award Year: 2023

ILLUMINATING MICROBIAL COMMUNICATION NETWORKS: THE PHYCOSPHERE LAB

Christophe COUDRET
CNRS & Toulouse University, France

Idan Tuval[1]; Glen Wheeler[2]; Jean-Baptiste Raina[3]

[1]CSIC-Illes Balears University; [2]Marine Biological Association of the United Kingdom (MBA); [3]UT Sydney

ABSTRACT
Marine microbial communities play a central role in global carbon and nutrient cycling. In these complex communities, chemically mediated signaling between phytoplankton and their neighboring microbes mediates important trophic interactions. Indeed, chemical signaling is the language of life at sea, and has long been assumed to be the best way for microorganisms to interact (from symbiotic to parasitic relationships). However, chemicals can only diffuse over very short distances. Are microorganisms also able to take long distance calls? Light, by its qualities (i.e., colors, intensity), and by its fluctuations (time, space), could be used for such a purpose. Most planktonic microorganisms are light-responsive (i.e., phototrophs, parasites), and can alter the local light seascape by scattering, absorption, and remission, or even by bioluminescence. Recent studies suggest that light-based communication may be an overlooked and potentially important avenue for microbial intra- and inter-species communication, influencing community structure and function in the ocean. Our project will conduct the first controlled experiments to assess the role of light communications using novel light-emitting nanoparticles) within and between microbial species. If microorganisms use light in addition to chemical signaling, the consequence of our research will be analogous to the discovery of a new language. Specifically, we aim to answer: What forms of light-mediated communication exist between microbial communities (i.e., chlorophyll fluorescence, bioluminescence)? Are bacteria able to sense light generated from their phytoplankton hosts? What are their physiological responses to light sensing (i.e., phototaxis or secretion of nutrients and infochemicals)?

KEYWORDS
Microbial ecology, Diatoms, Biophysics, Microfluidics, Luminescence

HFSP Reference Number: RGP007/2024
HFSP Award Category: Research Grant Program
HFSP Award Year: 2024

RESURRECTING THE ORIGINS OF TYROSINE KINASE ACTIVITY

Pau Creixell
University of Cambridge, UK

Brian Metzger[1]; Arun Shukla[2]
[1]Purdue University; [2]Indian Institute of Technology Kanpur

ABSTRACT
A central challenge in evolutionary biology is identifying the mechanisms that generate novel biological functions, a goal that can be best explored using tractable molecular systems. Protein tyrosine kinases represent an ideal model for this question: they are experimentally tractable, and yet, the origins of their specific activity remain poorly understood. While all organisms can use phosphorylation of serine and threonine residues as part of their signaling repertoire, tyrosine phosphorylation is found primarily in eukaryotes and evolved later in history. To resolve the timing and molecular and structural basis of this functional transition, we are integrating ancestral sequence reconstruction with protein biochemistry and structural biology. Here, we will provide an update on our latest progress and how our results provide initial answers to address whether the evolution of new molecular functions in natural proteins is dictated by rigid biophysical constraints, historical contingency, or a landscape of diverse, exchangeable solutions.

KEYWORDS
Evolution; Protein Tyrosine Kinases; Ancestral Sequence Reconstruction; Systems Biochemistry; Structural Biology

HFSP Reference Number: RGEC27/2024
HFSP Award Category: Research Grant Early Career
HFSP Award Year: 2024

HOW DO CIRCADIAN TEMPERATURE OSCILLATIONS BENEFIT THE HEALTH AND LONGEVITY OF C. ELEGANS?

Kristin Dahl
Basel University, Switzerland

Anne Spang
Basel University

ABSTRACT
We are all subject to changing temperatures throughout the year and even throughout the day. With the looming threat of climate change, it has become crucial to understand how living systems respond to temperature fluctuations. Caenorhabditis elegans, for example, are poikilotherms that naturally reside in soil and are therefore exposed to daily temperature changes within that environment. Because C. elegans cannot regulate their internal body temperature, their entire body has to respond to temperature changes. We sought to determine the effect of daily temperature changes, in the form of oscillations that recapitulate diurnal temperature gradients. Temperature oscillations are physiologically relevant, as they mimic environmental conditions that C. elegans may experience in their natural soil habitat. Surprisingly, we found that C. elegans exposed to temperature oscillations ranging from 13°C to 23°C, at a rate of 1°C per hour over a 24-hour period, exhibited an increased average median survival compared to animals maintained at constant temperatures of 23°C, 20°C, or 18°C. While either acute heat shock condition or maintenance at 13°C increased longevity, these conditions were associated with a reduction in progeny. In contrast, temperature oscillations increased lifespan without reducing reproductive fitness. Thus, circadian temperature oscillations provide a benefit to longevity without a cost to reproductive output. We hypothesized that the benefit conferred by oscillating temperatures was due to the induction of a mild stress response. To test this, we examined expression of hsp-4 as a readout of the UPRER and found that hsp-4 expression was modestly increased under oscillating temperatures compared to all steady-state conditions after two days. When temperature treatments were extended to seven days and animals were subsequently challenged with tunicamycin, those in the oscillating condition mounted a stronger response, indicating an extended capacity to respond to stress. Together, these data demonstrate that our temperature oscillation paradigm confers benefits to both lifespan and healthspan possibly through ER stress response pathways. This study highlights the adaptability of C. elegans, and we plan to continue by analysing the molecular regulators of this process, with a particular focus on neuronal mechanisms.

KEYWORDS
Stress response, Aging, Neuron adaptation, Proteomics, Thermal stress

HFSP Reference Number: LT0035/2024-L
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HFSP Award Year: 2024

BLUE IRIDESCENCE COEVOLVES WITH COLOUR VISION AND FLIGHT IN MORPHO BUTTERFLIES

Vincent Debat
Muséum National d'Histoire Naturelle (MNHN), France

Marco Giraldo[1]; Adriana Briscoe[2]; Florian, Titus Muijres[3]; Violaine Llaurens[4]; Gregor Belušic[5]
[1]Antioquia University; [2]The Regents of the University of California, Irvine; [3]University of Wageningen; [4]College de France; [5]University of Ljubljana

ABSTRACT
Understanding trait evolution is challenging because traits do not evolve in isolation, but rather in interaction with other traits. How these interactions shape phenotypic adaptation is a central question in evolutionary biology. This project focuses on Morpho, a genus of Neotropical butterflies famous for the blue iridescence of some species. The evolutionary drivers of this spectacular coloration remain elusive. We propose that blue iridescence evolved in tight connection with colour vision and flight. Coevolution with vision may result from selection on species recognition and mate choice, while coevolution with flight may be driven by predation, as bright flashes produced during flight could enhance escape success. We first quantified iridescence across the genus and evaluated phylogenetic constraints on its evolution. Although phylogeny influences patterns of variation, marked divergence occurs among species, contrasting pigmentary and diffuse iridescent species with highly directional bright blue species. We then investigated the evolution of the visual system by analysing opsin sequences and spatial expression, retinal organization, and photoreceptor spectral sensitivity. Morpho visual system (five opsins (UV, B, and three LW), at least six ommatidial types), suggests fine tuning of blue sensitivity: notably, expression of a blue-shifted green opsin in R1–R2 photoreceptors enables dense sampling of the blue–green spectrum. Opsin sequences show signatures of positive selection associated with both wing coloration and light environment. High polarization sensitivity endows some species with the ability to detect polarized signatures of iridescent wings. Next, we characterized escape flight behaviour in the wild using a videographic system. During simulated attacks, M. menelaus markedly increases flight erraticity, zigzagging horizontally at the cost of reduced speed. This unpredictability likely synergizes with flash coloration, enhancing escape probability. Analyses of codivergence between flight and iridescence are ongoing, and preliminary results will be presented. Experiments using an artificial light setup mimicking Morpho reflectance further show that wing beat frequency strongly enhances visual attraction in M. helenor, supporting adaptive coevolution between flight and iridescence.

Overall, our results indicate that iridescence jointly coevolved with vision and flight, shaped by sexual selection (species recognition and mate choice) and predation. Because Morpho species fly at different heights and times of day, they experience distinct light environments. We are currently comparing two sister species, M. eugenia and M. marcus, which differ in wing coloration and flight time but show similar visual performance. This comparison tests whether stabilizing selection on visual systems may have promoted divergent wing coloration, consistent with the sensory drive hypothesis.

KEYWORDS
Morphological evolution, iridescence, vision, flight, butterflies

HFSP Reference Number: RGP005/2023
HFSP Award Category: Research Grant
HFSP Award Year: 2023

ASSEMBLY, MECHANICS AND GROWTH OF PLANT CELL WALLS

Pauline Durand-Smet
Paris Cité, University France

Cyril Grandjean[1]; Ravi Shanker[2]; Anran Mao[2]; Sarah A. Pfaff[3]; Jordi Chan[4]; Sophie Asnacios[1]; Atef Asnacios[1]; Sulin Zhang[3]; Daniel J. Cosgrove[3]; Enrico Coen[4]; Anna J. Svagan[2]
[1]Paris Cité University; [2]KTH Royal Institute of Technology; [3]Pennsylvania State University; [4]John Innes Centre

ABSTRACT
Fibre-based materials support bridges, clothe us, coat our pills, seal our wounds, sustain every tree and shrub, and provide animals with their backbones, exoskeletons and muscles. Synthetic fibre-based materials are shaped by design, but how biological fibre networks self-organise to generate shapes with their associated mechanical properties remains an open question. We address this problem by using plant cells as a model system. Plant cells are immotile, which simplifies analysis, and their mechanical behavior relies mostly on the cell wall—a fibre-based composite structure built primarily from cellulose, hemicellulose, and pectin. We make use of plant protoplasts - single cells enzymatically deprived of their cell walls, yet capable of wall regeneration. To dissect the feedback between shape and cellular self-organization, we first imposed shapes on regenerating plant cells by confining them into microwells of specific geometry. We show that plant cells can grow and divide within these microwells, and the geometry influences the cell polarity and fibers organization. Upon release from the microwells, cells retain a “shape memory” enabling future analysis of how subsequent growth may override the imposed shapes. To understand the mechanical basis underlying shape memory, our team compared the structure and mechanical properties of regenerating cell walls in plant protoplasts with those of a synthetic wall analogue. We show that assemblies with increasing numbers of layers of cellulose nanofibers and pectin recapitulate the increase in stiffness observed in natural walls during thickening. Thus, basic wall mechanics emerge from simple material combinations. Building on this foundation, we are now developing a quantitative understanding of how intra- and extracellular fibre networks interact to control cell growth and division based on our approach combining live-cell imaging, computational modelling, and micro-engineering.

KEYWORDS
plant cell wall, mechanics, fiber network, polarity, synthetic walls

HFSP Reference Number: RGP0005/2022
HFSP Award Category: Research Grant Program
HFSP Award Year: 2022

ARE CENTRIOLES TRULY CENTRAL? PROBING THE EVOLUTION OF CELL SKELETON ORGANISING CENTRES USING A UNIQUE DEEP-BRANCHING PROTIST

Yana Eglit
University of Geneva, Switzerland

Virginie Hamel ; Paul Guichard
University of Geneva

ABSTRACT
In animal cells, centrosomes are integral to cell division and contain at their core a pair of barrel-shaped microtubule structures, the centrioles. In most cell types, centrioles, in the form of 'basal bodies', constitute the basis for a flagellum, a whip-like structure that is often motile and may provide propulsion for the cell. Protists, or eukaryotes outside the classical Kingdoms Plants, Animals, and Fungi, are predominantly unicellular microbes and in fact comprise most of the eukaryotic diversity. Many protists are flagellates, usually carrying two flagella whose basal bodies often lie at the focal point of the cell skeleton. Current evolutionary analyses strongly suggest that the last common ancestor of all eukaryotes ('LECA') was also a flagellate, presumably with a fully formed basal body. Throughout eukaryotes, numerous lineages exist that lack flagella and centrioles entirely, presumably through loss. In some of these organisms, there are distinctive structures at the focal point of the cell skeleton, called a MicroTubule Organising Centre (MTOC). Several types of these structures have evolved independently of each other; and very little is known about their evolutionary origin. To investigate whether some of these structures may be highly reduced relics of an ancestral centriole, we are examining the peculiar MTOCs of the distinctive non-flagellate protist Meteora sporadica by combining super-resolution imaging techniques with sequence-based bioinformatic searches for potential homologues of key centriolar proteins. We expect that our findings will shed light on not only the origin of non-centriolar MTOCs among protists, but also on the evolution of the animal centrosome itself.

KEYWORDS
evolutionary cell biology, protists, cytoskeleton, super-resolution microscopy

HFSP Reference Number: LT0037/2025-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2025

FROM NANO TO ORGANISMAL SCALE: STRUCTURAL REGULATION OF REGENERATING JELLYFISH

Daniel Font
Observatoire Océanologique de Banyuls-sur-Mer, CNRS/University of Sorbonne, France

Anne Materne[2]; Carl Modes[2]; Ulyana Shimanovich[3]; Chiara Sinigaglia[1]
[1]Observatoire Océanologique de Banyuls-sur-Mer, CNRS/University of Sorbonne; [2] Max Planck Institute of Molecular Cell Biology and Genetics (MPI-CBG); [3] Weizmann Institute of Science

ABSTRACT
The ability of organisms to regenerate after being injured, or their regenerative potential, varies among metazoans. The extracellular matrix (ECM), which influences progenitor cell behaviour and contributes to tissue mechanical characteristics, is a common player in the repair processes. We are using our laboratory jellyfish model, Clytia hemisphaerica, to examine tissue healing and morphogenesis from a novel perspective. The jellyfish form has a very thick, highly hydrated extracellular matrix called mesoglea. Although the mesoglea undergoes significant remodelling during the quick healing process (the umbrella form is recovered within 12 hours of damage), it is still unknown how much it influences the dynamics of the tissues that surround it and, eventually, the restoration of the overall shape of the organism. The mesoglea layer's relative thickness places restrictions on the animal body's ability to flex and reshape. The jellyfish umbrella is covered by a layer of smooth muscle-epithelial cells. As the jellyfish undergoes remodelling, distinct +1 topological defects appear in the arrays of smooth muscle fibres. Progenitors proliferation and the morphogenesis of new organs take place in these zones, which correlate to areas of stem cell recruitment into a regenerative blastema. Although it is still unknown how the +1 topological defects relate to the ECM features, we hypothesise that they may serve as stable hubs to promote morphogenesis. To find out how the mesoglea's material properties, which are still unknown, affect the repair process, we are employing a multidisciplinary approach. We have devised a technique to separate the mesoglea from the jellyfish tissues and we are manipulating mesoglea remodelling with different drugs, which target either the processing of new collagen or its degradation. We are characterizing how the mesoglea's composition, structure, and mechanical qualities are altered throughout the repair process; these data is informing the generation of an in-silico model of the animal (Modes' lab) that considers dynamic organismal shape and links the mesogleal characteristics to theoretical topological defect mechanics.

The data will further guide the synthesis of synthetic replica of mesoglea (Shimanovich's lab). By combining in vivo, in vitro and in silico methods, we will be able to investigate the dynamic interaction between ECM and morphogenesis in new ways and discover underlying principles that connect events in shape restoration from the cellular to organismal levels in metazoans.

KEYWORDS
extracellular matrix, regeneration, jellyfish, topological defect

HFSP Reference Number: RGP020/2024
HFSP Award Category: Research Grant
HFSP Award Year: 2024

TOWARDS THE RECONSTITUTION OF GEOBACTER SPLIT PILIN ASSEMBLY AND NANOWIRE SECRETION IN HETEROLOGOUS HOSTS

Olivera Francetic
Institut Pasteur, France

Nikhil Malvankar
Yale University

ABSTRACT
Bacterial anaerobic respiration in the absence of soluble electron acceptors requires extracellular electron transfer (EET) via protein nanowires to minerals or partner cells. EET drives various global environmental phenomena and bioremediation, bioenergy, and bioelectronics. However, the composition and structure of these nanowires and the molecular basis of their conductivity were unknown. Since 2002, it has been universally accepted that nanowires are a Type 4 pilus system (T4P) made up of an unusually short pilin subunit PilA-N. However, our genetic, biophysical, and biochemical data challenged this 20-year model (1). We found that Geobacter sulfurreducens (Gs) PilA-N associates with another protein, PilA-C, to form PilA-NC filaments that remain intracellular during EET. Both PilA-N and PilA-C are required for the secretion of polymerized cytochromes that are necessary for EET. As in the dictum by Richard Feynman, "What I cannot create, I do not understand," successful design is a powerful way to show that a design principle has been understood. Here, we aimed to achieve this goal by reconstituting the assembly of PilA-N, PilA-C into pili in Escherichia coli (Ec), and by identifying their assembly machinery and their role in the secretion of nanowire-forming cytochromes OmcS and OmcZ. To assemble PilA-NC pili in Ec, we used the Ec T4P assembly system (T4PS) and the Klebsiella oxytoca (Ko) Type-2 secretion system (T2SS). Both systems, despite being fully functional, fail to assemble Gs pili, contradicting several prior reports of PilA-N assembly in heterologous hosts, despite using the nearly identical T4PS. This is mainly due to the failure of prepilin peptidases from these systems (GspO and PulO) to promote proteolytic maturation of PilA-N. Partial PilAN maturation in Ec was achieved by the Gs prepilin peptidase PilD. We therefore reconstituted one of the Gs T4P assembly systems encoded by several gene clusters via Gibson assembly. Deleting the pilT4-pilS-pilR genes led to increased expression of downstream genes pilAN-pilAC. The Gs system reconstituted in Ec allowed assembly of PilA-N pili in the presence of PilA-C in select strains under specific culture conditions. We also cloned in Ec the complete Gs T2SS gene cluster, the omcS (hereafter, osc) and OmcZ nanowire assembly cluster with respective isomerases and proteases. OmcS protein was expressed in the presence of this cluster but not on its own, suggesting that it is prone to degradation. However, the osc gene cluster was not sufficient to assemble OmcS nanowires on the Ec cell surface. The OmcZ did not fold properly due to inefficient heme incorporation, as evidenced by protein aggregation even in the presence of Ec heme incorporation genes. Current efforts are aimed at overcoming these issues by testing Gs heme incorporation genes or anaerobic growth conditions.

We are complementing these studies in Gs by deleting and complementing pilA-N and pilA-C, along with their point mutants, to understand pilus assembly and nanowire secretion mechanism.

1. Y. Gu et al., Structure of Geobacter pili reveals secretory rather than nanowire behavior. Nature 597, 430-434 (2021).

KEYWORDS
extracellular electron trasfer, environmentally important bacteria, cryo-electron microscopy and tomography, atomic and electrostatic force microscpoy, cytochrome secretion system

HFSP Reference Number: RGP017/2023
HFSP Award Category: Research Grant Program
HFSP Award Year: 2023

EXPERIMENTALLY EVOLVING CELLULAR MINIATURIZATION

Marco Fumasoni
Gulbenkian Institute of Molecular Medicine (GIMM), Portugal

Ana Garoña[1]; Morgane Lemos[1]; Andrea Giometto[2]
[1]Gulbenkian Institute of Molecular Medicine (GIMM); [2]Cornell University

ABSTRACT
Cell volume, a key determinant of physiology, is maintained by cell size homeostasis. Large deviations from typical size are often harmful, yet cell sizes have diverged drastically in evolution. How does size homeostasis evolve to support such diversity without impairing physiology? To address this, we used experimental evolution to select progressively smaller Saccharomyces cerevisiae cells. Over 1,500 generations, we achieved 4 to 6-fold volume reductions compared to wild type. Size homeostasis remained robust and evolutionary stable, with cells maintaining competitive fitness, indicating that dramatic volume changes need not compromise physiology. We investigated the genetic basis of cellular miniaturization through whole-genome sequencing of the evolving populations. We show that genetic manipulations of the G1 cyclin CLN3 and the Greatwall kinase RIM15 signaling cascades produce large changes in cell size, with adaptive mutations recapitulating the evolved volumes and loss-of-function mutations yielding enlarged cells. Our results demonstrate the evolutionary plasticity of cell size homeostasis and reveal a mechanism for eukaryotic cell size evolution.

KEYWORDS
Cell size, Experimental Evolution, Cell cycle, Intracellular scaling, S. cerevisiae

HFSP Reference Number: RGEC28/2023
HFSP Award Category: Research Grant Early Career
HFSP Award Year: 2023

ADVANCED MICROSCOPY AND INTEGRATIVE MODELLING TO RESOLVE THE STRUCTURAL DYNAMICS AND BIOPHYSICS OF EXOCYTOSIS

Oriol Gallego
Pompeu Fabra University, Spain

Sasha Meek; Marta Puig-Tintó; Sebastian Ortiz; Sebastien Tosi; Alex De Marco; Daniel Castaño-Diez

ABSTRACT
State-of-the-art: The exocyst is the protein complex that tethers secretory vesicles to the plasma membrane in constitutive exocytosis, an essential process that delivers biomolecules to the surface of all eukaryotes. Importantly, the tethering of a single vesicle involves multiple copies of the complex, implying that their activity must be somehow concerted. Unfortunately, none of the available microscopy techniques have been capable of capturing such dynamic and convoluted machinery.
Scientific question: What, then, is the exocyst higher-order structure (ExHOS) that coordinates the action of these multiple exocysts? What are the conformational switches that underly its function? Are these structural kinetics adapted to suit the optimal physical chemistry of organisms that grow within different environments?
Results: With the support of HFSP we integrated particle tracking, super-resolution microscopy and cryo-electron tomography to time-resolve the continuum conformational landscape of the ExHOS and to functionally annotate its different conformations. We found that 7 exocysts form flexible ring-shaped ExHOS that tether vesicles at < 45 nm from the plasma membrane. The ExHOS rapidly expands while pulling the vesicle towards the plasma membrane in a stepwise mechanism comprising three metastable states at 27, 18 and 5 nm from the plasma membrane. These results were recently published (Puig-Tintó et al., 2026, Cell). After resolving the spatiotemporal dynamics of the ExHOS, we further resolved the conformational kinetics that underly the different metastable states of exocytosis. We are now continuing the investigation into how these kinetics scale with temperature and have found that the sub-steps of the process have differential behaviors.
Significance beyond this study: By revealing fundamental insight on the mechanism of exocytosis, this work illustrates how multimodal imaging and integrative modelling can open new avenues to investigate the cell's machinery. We have now postulated the biophysical principles that constrain the adaptation of the exocytic machinery, hoping that this might help us to understand the evolution of organisms that grow under different temperatures.

References:
https://doi.org/10.1016/j.cell.2025.11.038

KEYWORDS
Exocytosis, Super resolution microscopy, cryo-Correlative Light-Electron Microscopy, Integrative modelling, Biophysics

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HFSP Award Category: Research Grant Program
HFSP Award Year: 2020

THE ARCHITECTURE OF PHOTOSYNTHESIS

Anja Geitmann
McGill University, Canada

Craig Brodersen[2]; John Dear[3]; Matteo Pezzulla[4]
[1]Yale University; [2]Imperial College of Science, Technology and Medicine; [3]Aarhus University

ABSTRACT
Photosynthesis is the metabolic process that sustains all life on Earth. Understanding how vascular plants reconcile leaf structural optimization for both light capture and photosynthetic gas exchange with architectural stability has been an intractable challenge. While the anatomy of the inner leaf tissues should optimize gas exchange through a porous microstructure that augments the net absorptive interface, such an optimization should also come with a cost to the architectural stability of thin planar leaves. We leverage a combination of cell biology, ecophysiology, and mechanical engineering to investigate the functional trade-off between gas exchange and leaf mechanics. Based on engineering concepts, the biomechanics of plant leaves and the porous properties of its inner tissues are analyzed at multiple length scales to gain insight into evolutionary pressures that have shaped laminar leaves over millions of years. Our approach will reconcile how and why leaves are structured the way they are and identify the critical design principles of highly productive species capable of sustaining maximum photosynthetic performance. We also aim to understand the overall construction of the leaf, in terms of its sandwich structure of epidermis and mesophyll, as well as the role of turgor pressure, to produce a laminar leaf that is biomechanically robust whilst meeting the demands to serve as the cellular scaffold for photosynthesis.

KEYWORDS
plant science, biomechanics, photosynthesis

HFSP Reference Number: RGP010/2023
HFSP Award Category: Research Grant
HFSP Award Year: 2023

MOSQUITO-BORNE HUMAN PATHOGENIC VIRUSES USE FUNDAMENTALLY DIFFERENT MEMBRANE INTERACTIONS IN INSECT VERSUS MAMMALIAN CELLS

Gisa Gerold
Medical University of Innsbruck, Austria

Anja De Bruin[1]; Lathika Valliyott[2]; Enzo Mameli[3]; Jesse S Plung[3]; Andres Merits[4]; Jonathan Abraham[3]; Norbert Perrimon[3]; Nick Ariotti[2]
[1]Medical University Innsbruck; [2]The University of Queensland ; [3]Harvard Medical School; [4]University of Tartu

ABSTRACT
Insect cell membranes differ from mammalian membranes by deformability, lipid content and distribution. Enveloped insect-borne viruses such as the alphavirus chikungunya virus (CHIKV) require intimate interactions with cellular membranes to enter cells, replicate their genomes in cells and bud from cells. Despite fundamental biophysical differences between insect and mammalian membranes, viruses can productively infect cells from both phyla. Decades of studies on insect-borne viruses have not addressed the machinery of insect membrane deformation and its exploitation by viruses. Working at the interface of insect genetics, biophysics and infection biology, we have combined innovative technologies to break through this barrier in the field. Specifically, we: 1. Employed gene editing libraries in insect cells and identified an unexpected receptor molecule for virus binding and uptake; and 2. Imaged and characterized virus-induced membrane deformation at high resolution in insect versus mammalian cells. Using a suicide gene modified mosquito-borne CHIKV and an insect cell-based CRISPR loss-of function screen, we discovered a CHIKV insect cell receptor and multiple enzymes involved in its biosynthesis pathway. By RNA interference in insect cells and overexpression of the insect receptor in refractory mammalian cells, we confirm that the protein is a CHIKV host factor. We show that CHIKV E2–E1 glycoproteins directly bind the receptor, facilitating virus attachment and internalization. We further show that the receptor rendered cells susceptible to other arthritogenic alphaviruses including Semliki Forest virus (SFV) and Ross River virus (RRV), while the encephalitic alphavirus Venezuelan equine encephalitis virus (VEEV) could not enter complemented cells. Interestingly, while the ectodomains of the insect and the mammalian receptors show similarities, their membrane anchors are fundamentally different. Similarly, we found differences in the architechture and localization of membrane microdomains, which CHIKV uses for replicating its RNA genome. In mammalian cells, replication complexes, termed spherules, localized to the plasma membrane with defined architecture and associated rearrangement of cellular organelles. In contrast, we could not detect any deformation of the plasma and instead observed intracellular protein arrays in CHIKV replicating insect cells. Using lipid probes and split APEX approaches, we now aim to uncover the lipid profile of virus-induced microdomains in both species and understand organelle re-arrangements, respectively.

Altogether, we identify a mosquito receptor for CHIKV entry and demonstrate that mosquito-borne viruses have evolved to switch between distinct mechanisms for entry and replication in insect versus mammalian cells.

KEYWORDS
Membrane Biology; Insect Cells; Mosquito-borne Viruses; Receptor; Replication Complex

HFSP Reference Number: RGP011/2023
HFSP Award Category: Research Grant Program
HFSP Award Year: 2023

OXYGEN ISOTOPES OF METHANOL LINK DARK OXYGEN PRODUCTION AND METHANE OXIDATION IN GROUNDWATER

Tetyana Gilevska
The University of Southern Denmark, Denmark

Toshiki Nagakura[1]; S. Emil Ruff[2]; Jordon D. Hemingway[3]; Carolina Carvalho[3]; Emeline Vidal[2]; Beate Kraft[1]
[1]University of Southern Denmark (SDU); [2]The Marine Biological Laboratory; [3]ETH Zürich

ABSTRACT
Microbial dark oxygen production (DOP) may provide an overlooked oxygen source sustaining methane oxidation in groundwater. Methanol (MeOH), the first intermediate of aerobic methane oxidation, is a sensitive tracer of the oxygen source because its oxygen atom is directly derived from the oxidant and does not exchange with water. Here, we tested whether oxygen incorporation into methanol can be traced isotopically under controlled conditions. Pure-culture incubations of the methanotroph Methylosinus sporium were performed using $^{18}O_2$. To enable detection of this transient intermediate, methanol consumption was inhibited, allowing MeOH to accumulate. A salting-out headspace GC–MS method enabled quantification of MeOH and ^{18}O-MeOH at low micromolar concentrations. The formation of ^{18}O-labeled methanol directly from $^{18}O_2$ confirms oxygen incorporation during methane oxidation and validates methanol oxygen isotopes as a tracer. This experimental framework establishes the basis for future co-culture experiments combining methanotrophs with oxygen-dismutating microbial partners to detect and trace DOP-derived oxygen supporting methane oxidation.

KEYWORDS
Dark Oxygen Production, Methane Oxidation, Methanol Isotopes, ^{18}O-labeling, Methanotrophs, GC–MS

HFSP Reference Number: RGEC34/2023
HFSP Award Category: Research Grant Early Career
HFSP Award Year: 2023

MORPHOLOGICAL BASES OF BLUE IRIDESCENCE DIVERSITY ACROSS MORPHO BUTTERFLIES

Marco Giraldo
University of Antioquia, Colombia

Mariana Jaramillo[1]; Juliana Sosa[1]; Ana Valencia[1]; Melanny Silva[1]; Vincent Debat[2]; Gregor Belusic[3]
[1]University of Antioquia; [2]Muséum National d'Histoire Naturelle (MNHN); [3]University of Ljubljana

ABSTRACT
Morpho butterflies are renowned for their brilliant structural coloration, classically attributed to the "Christmas tree" ridge architecture of their dorsal wing scales. However, across the genus, wing coloration varies markedly in hue, intensity, angular distribution, and polarization properties. This diversity raises the question of whether variation of the canonical structural model adequately explains the optical variability observed among Morpho species. To examine how nanostructural variation shapes optical output, we studied the thirty species of the Morpho genus, by combining spectrophotometry, scatterometry, polarization-resolved reflectance measurements, scanning and transmission electron microscopy (SEM and TEM), and finite-difference time-domain (FDTD) simulations based on experimentally-derived geometries. Morphological analyses reveal substantial interspecific differences in ridge spacing, lamellar thickness, multilayer organization, ridge inclination, and degree of structural disorder. In several species, the scale architecture departs from the classical Christmas tree configuration. These structural differences correlate with pronounced variation in spectral reflectance and polarization signatures. Although all species exhibit coloration arising from multilayer interference within the ridges, the strength and spectral distribution of polarization-dependent reflectance vary considerably. FDTD simulations reproduce the principal spectral and polarization features and show that small changes in nanostructural parameters, particularly lamellar periodicity, ridge separation and thickness, can generate distinct optical responses. Lastly, the reflectance of the wings strongly depends on the spectral properties of the natural illuminant, e.g. morning skylight, daylight, or light in the green understory. We show extensive variation across species in the coloration mechanisms that create robust patterns of reflectance, i.e. that are largely independent of the illuminant type, or patterns that change with the illuminant, which might affect camouflage or intraspecific recognition. Overall, our results indicate that Morpho wing coloration is not based on a single shared architectural organization, but on a structurally diverse panel of traits in which subtle morphological variations produce species-specific spectral and polarization outcomes.

KEYWORDS
butterfly coloration, multilayers, FDTD, iridescence, polarization

HFSP Reference Number: RGP005/2023
HFSP Award Category: Research Grant
HFSP Award Year: 2023

MULTI-ENZYME MOF NANOREACTORS TRANSPLANT A DRUG BIOSYNTHESIS PATHWAY INTO CELLS

Raik Grünberg
King Abdullah University for Science and Technology (KAUST), Saudi Arabia

Ainur Sharip; Somayah Qutub; Manar Farooqui; Walaa Baslyman; Nida Khalfay; Lukman Alimi; Patricia Lopez Sanchez; Lingyun Zhao; Milena Chernyshevskaia; Giovanni Colombo; Niveen Khashab; Stefan Arold; Raik Grünberg
King Abdullah University for Science and Technology (KAUST)

ABSTRACT
Most proteins evolved to function within multi-protein systems such as metabolic networks or signaling pathways. Our technical applications of proteins, by contrast, are mostly constrained by a single protein paradigm. Reliable methods for the stabilization, in vitro operation and intracellular delivery of integrated multi-protein systems could therefore open up new ways to interface with and manipulate biological systems, with applications ranging from biotechnology to therapy. We demonstrate that the entire violacein biosynthesis pathway, consisting of up to six separately purified enzymes, can be infiltrated into a hierarchically etched MIL-101 (eMIL) metal organic framework. The eMIL micro-environment reshaped pathway dynamics and reaction flows, multiplied product yield and enabled pathway reuse and storage. More importantly, eMIL nanoreactors delivered the six-protein system into mammalian cells, where it produced the bacterial natural product and drug candidate violacein from cell-provided substrates and cofactors. To date, this represents the most complex protein system delivered into cells. We believe that violacein nanoreactors will pave the way towards a novel class of intracellular protein systems therapies.

KEYWORDS
protein delivery, biosynthesis pathways, multi-protein systems, protein therapeutics

HFSP Reference Number: LT00334/2007-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2007

CROSS-TALK BETWEEN NON-CANONICAL WNT AND TRANSFORMING GROWTH FACTOR BETA PATHWAYS CONTROL THE EPITHELIAL-MESENCHYMAL TRANSITION AND CELL MIGRATION

Taran Gujral
Fred Hutch Cancer Center, USA

ABSTRACT
The epithelial-mesenchymal transition (EMT) is a central mechanism for diversifying the cells found in complex tissues. During EMT, epithelial cells adopt mesenchymal properties by altering their morphology, cellular architecture, adhesion, and migratory capacity. This dynamic process helps organize the formation of the body plan, and EMT is well studied in the context of embryonic development including mesoderm and neural crest formation. Previously, we discovered a new non-canonical Wnt5-Frizzled 2 (Fzd2) pathway that drives EMT and mediates cell migration. Our follow-up work uncovered that EMT depends on co-operation between two developmentally important pathways – Wnt5/Fzd2 and Transforming growth factor beta (TGFb). We found that Fzd2-driven EMT is associated with upregulation of TGFb receptor levels and subsequent activation of TGFb pathway. Similarly, TGFb-driven EMT is accompanied by increased expression of Wnt5a and Fzd2. Together with our observations that expression of Fzd2 turns mesenchymal cells hyper-sensitive to TGFb stimulation, whereas Fzd2 inhibition activates a novel compensatory response that also ensures sustained TGFb signaling, this suggests a complex a co-regulatory interaction between these two evolutionary conserved pathways. Importantly, inhibition of both Fzd2 and TGFb pathway completely abrogates the key feature of EMT, the mesenchymal cell migration. Thus, we propose that EMT is driven by a robust co-regulatory mechanism that depends on both Fzd2 and TGFb signaling to ensure a complete transdifferentiation.

KEYWORDS
Wnt, TGFb, developmental signaling, networks, EMT

HFSP Reference Number: LTF0584/2009
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2009

ENHANCED BASELINE CYTOTOXIC T CELL FUNCTION PREDICTS RESPONSE TO PD-1 BLOCKADE IN RICHTER TRANSFORMATION

Johan Gustafsson
University of Gothenburg, Sweden

Chiara Tomasoni[1]; Graydon Moorhead[2]; Gabriela Brunsting Hoffmann[3]; Erik Landolsi[4]; Wesley Lu[3]; Ruitong Li[2]; Shuqiang Li[3]; Nicholas Haradhvala[2]; Connor Johnson[2]; Stephanie Deng[3]; Donna Neuberg[3]; Xia Bu[3]; Gordon J Freeman[3]; Othman Al-Sawaf[5]; Barbara Eichhorst[5]; Erin Parry[3]; Gad Getz[2]; Catherine Wu[3]; Elisa Ten Hacken[6]

[1]Weill Cornell Medicine; [2]Broad Institute of MIT & Harvard; [3]Dana Farber Cancer Institute; [4]Chalmers University of Technology; [5]University of Cologne; [6]Cornell University

ABSTRACT
Chronic lymphocytic leukemia (CLL) is the most common adult leukemia in Western countries and is typically an indolent B-cell malignancy. Many patients achieve long-term disease control with targeted therapies, such as BTK and BCL2 inhibitors. In contrast, Richter transformation (RT), in which CLL progresses into an aggressive lymphoma, is associated with rapid clinical deterioration, limited treatment options, and poor prognosis. There is therefore a strong need to understand RT biology and identify effective therapies. Immune checkpoint blockade (ICB) has shown activity in RT, but the tumor microenvironment (TME) features governing response remain poorly defined. We combined single-cell and spatial transcriptomics, functional immunologic assays, and in vivo immunotherapy experiments to dissect the immune landscape of CLL and RT and identify determinants of ICB response. Single-cell RNA sequencing of splenocytes from 18 genetically diverse mouse models of CLL and RT revealed that RT is enriched in cytotoxic CD8+ and CD4+ T cells with activation and exhaustion signatures, pro-inflammatory macrophages, and heightened IFNγ signaling compared to CLL. Whole- genome sequencing demonstrated a higher mutational burden in RT, and computational prediction identified high-affinity neoantigen candidates, consistent with increased antigenic potential. Single-cell TCR sequencing revealed increased clonal expansion of RT T cells, supporting antigen-driven T-cell expansion. Functionally, RT T cells produced more granzyme B and inflammatory cytokines upon stimulation and effectively induced tumor-cell killing in co-culture assays, unlike CLL T cells. To test the role of IFNγ signaling, we performed secondary RT transplants in genetically modified mice lacking IFNγ receptor (i.e., IFNGRKO). Tumor growth was reduced compared to controls, indicating that IFNγ-driven TME remodeling promotes RT progression. To examine determinants of therapeutic response, four genetically distinct RT mouse models were treated with anti-PD-1 therapy, yielding responders and non-responders. Responders displayed reduced T-cell exhaustion signatures after treatment and improved ex vivo tumor cell killing by both CD4+ and CD8+ T cells, whereas non-responders retained dysfunctional T-cell states.

Notably, response to ICB was associated with preserved lymphoid architecture. Responder models maintained intact splenic structure with defined T-cell zones, greater T-cell clonal diversity, and markers of migratory capacity, whereas non-responders showed disrupted tissue organization. Extending these findings to patients, spatial transcriptomic and multi-immunofluorescence analyses of 12 pre-treatment lymph node biopsies from RT patients receiving anti-PD-1–based therapy are ongoing to determine whether similar architectural features and immune–tumor spatial relationships associate with clinical response in humans. Overall, our results indicate that preserved lymphoid architecture and pre-existing functional cytotoxic T cells are associated with response to ICB in RT, and that IFNγ signaling plays an important role in RT biology by promoting tumor-immune interactions. These findings will ultimately define why immunotherapy benefits only a subset of patients and may inform biomarker development and treatment strategies for this aggressive disease.

KEYWORDS
CLL, Richter Transformation, interferon gamma, tumor microenvironment, anti-PD1

HFSP Reference Number: LT0043/2023-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2023

DEFINITION AND MOLECULAR REGULATION OF AGING IN ORGANISMS WITH DIVERSE LIFESPANS AND LIFE CYCLES

Motoshi Hayano
Tokyo University Of Science, Japan

Manami Shibasaki[1]; Masa-Aki Yoshida[2]; Prof Shigeharu Kinoshita[3]
[1]ASAGI Labs; [2]Shimane University; [3]The University of Tokyo

ABSTRACT
The National Institute on Aging (NIA) states that "aging is accompanied by gradual changes in most body systems," highlighting that aging is a progressive physiological change over time, not necessarily synonymous with mere lifespan. In most mammals, physiological traits such as respiratory rate, heart rate, reproductive timing, and body size correlate with lifespan, and it is indisputable that aging remains the primary risk factor for various diseases. We have previously established the "ICE" (inducible changes to the epigenome) mouse model, demonstrating that epigenetic alterations driven by DNA damage—independent of genomic mutations—actively induce the aging process (Yang and Hayano et al., 2023 Cell). Aging fundamentally involves the loss of cell-type-specific gene expression patterns due to these epigenetic shifts. Indeed, phenomena such as "mesenchymal drift" (MD) suggest that aging and lifespan are actively determined in a species-specific manner (Lu et al., 2025 Cell). Building upon these findings, our current research investigates the molecular mechanisms of acquired aging not only in mammals, such as humans and mice, but also across organisms with diverse life cycles. We are analyzing reproduction-induced rapid aging in annual fish, such as the ayu, and the mechanisms underlying suicidal reproduction in octopuses. Furthermore, by conducting genomic analyses on extreme-longevity species like the Greenland shark (which boasts a lifespan of 400–500 years), we are comparing aging-regulating molecules across taxa. Through these comparative studies, we aim to redefine aging and elucidate its molecular control across organisms with diverse lifespans and life cycles. In this context, we wish to discuss the dynamics of various regulatory factors, ranging from growth pathways like FOXO and mTOR to immune and epigenetic regulators.

KEYWORDS
aging, diverse lifespans, epigenome, mammal, fish

HFSP Reference Number: HFSP2019-I/530
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2019

CONSERVED AND LINEAGE-SPECIFIC MECHANISMS DRIVE CHROMATOPHORE DIFFERENTIATION IN REPTILES

Pierre-Yves Helleboid
University of Geneva, Switzerland

Athanasia Tzika
University of Geneva

ABSTRACT
Pigmented cells have been extensively studied in teleost fishes, particularly zebrafish, where interactions among brown melanophores, yellow-to-red xanthophores, and reflective iridophores generate color patterns. These chromatophore types are also conserved in squamates, which diverged from teleosts approximately 400 million years ago. Squamates display a wide diversity of hues and patterns; however, unlike teleosts, they do not undergo developmental metamorphosis. Thus, they provide a valuable model to explore the evolution and development of vertebrate pigment cell diversity. We used single-cell transcriptomics to reconstruct the development of chromatophore lineages in two reptilian model species, the corn snake and the bearded dragon. In the corn snake, the differentiation of chromatophores resembles that of zebrafish chromatophores. In the bearded dragon, we uncovered two previously undescribed chromatophore populations related to melanophores and xanthophores that share a common transcriptional program and contribute to regional patterning. Furthermore, ASIP, a gene implicated in countershading in mice and zebrafish, is likely to play a similar role in the bearded dragon, and is widely expressed by iridophores in the corn snake. In addition, we investigated the major role of chromatophore transcription factors in the differentiation process, and we could highlight their central and evolving role in tetrapod color pattern evolution. We then integrated our corn snake dataset with published zebrafish and amphibian single-cell datasets, revealing that while differentiated pigment cell lineages rely on conserved molecular actors, progenitor populations display greater transcriptional diversity.

KEYWORDS
Chromatophores, Transcriptomics, Development, Reptiles, Pigment

HFSP Reference Number: RGP0037/2022
HFSP Award Category: Research Grant
HFSP Award Year: 2022

DISENTANGLING MICROBIOME EFFECTS ON PLANT DEVELOPMENT AND LIFE HISTORY

Alejandra Hernandez Teran
University of California, Irvine, USA

Maria Rebolleda-Gomez
University of California, Irvine

ABSTRACT
It is increasingly clear that microbes influence host health and fitness. In particular, microbial communities affect developmental transitions such as flowering time in plants, life-history decisions that shape plant performance in both natural systems and managed agroecosystems. However, we still lack a mechanistic understanding of which microbes alter such traits, how they do so, and whether these effects are adaptive for the microbes or consequences of their metabolism. The central goal of this project to decipher the mechanisms underlying microbiome effects on plant development. To address this question, I combined top-down and bottom-up experimental approaches. Using Arabidopsis thaliana, we first implemented a directed evolution experiment of whole communities by inoculating plants with distinct microbial communities and serially transferring microbiomes associated with early or late-flowering plants. After four transfers, independently selected communities consistently accelerated flowering by up to 7 days relative to controls. Despite distinct starting inocula, selected lines converged toward similar early-flowering phenotypes, suggesting that host-mediated selection can drive compositional and/or functional convergence at the community level. Across transfers, we quantified plant fitness-related traits and generated 16S rRNA gene and metabolic profiles to characterize microbial ecological and functional trajectories. We are currently identifying taxa that diverge early during selection and testing whether their temporal dynamics predict variation in flowering time. Complementing this approach, we implemented a bottom-up strategy using 10-member synthetic consortia (SynComs) to disentangle whether developmental shifts are driven by individual strains or by simplified community-level functions. By systematically assembling defined consortia and manipulating their composition, we test whether flowering time can be altered by specific taxa, functional guilds, or emergent properties of microbial interactions. Finally, we integrate experimental data into demographic life-history models for monocarpic plants. Using classical frameworks that estimate fitness as lifetime reproductive output, we evaluate whether microbes can shift optimal flowering time and other life-history parameters, and whether such shifts can be predicted from microbial composition and functional profiles. This integration allows us to move from descriptive community changes to quantitative predictions of microbiome effects on plant fitness landscapes. Together, our results demonstrate that microbiomes can be shaped through host-mediated selection and rational assembly to generate predictable shifts in plant developmental timing.

By combining evolutionary experiments, synthetic ecology, and demographic modeling, this work provides a mechanistic and predictive framework to understand how microbial communities influence plant life-history strategies, highlighting their potential as targets for microbiome-based approaches to improve crop performance and resilience.

KEYWORDS
Plant microbiome, SynComs, Plant life history

HFSP Reference Number: LT0047/2023-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2023

HARMONY IN DYNAMICS - EXPLORING EMERGENT COOPERATION THROUGH PARTICIPATORY ART

Uri Hershberg
University of Haifa, Israel

Luciano Perondi[1]; Goni Shifron[2]; Hasan Ayaz[3]
[1]Iuav University of Venice; [2]Multi-disciplinary independent artist; [3]Drexel University

ABSTRACT
Harmony in dynamics is an interdisciplinary research collective that brings together expertise in design, neuroscience, complex systems, and participatory art. The collaboration integrates the work ofLuciano Perondi in interface design, graphic and game design, and user testing at IUAV; Hasan Ayaz in wearable neuroimaging, neuroergonomics, and human interaction research at Drexel University; Uri Hershberg in complex systems and theoretical biology at the University of Haifa; and the art practice of Goni Shifron, a multidisciplinary artist based in France. Together, this collaboration investigates how individual actions converge into adaptive group dynamics. We present a novel art-science research framework centered on the participatory artwork "Tä". Using methods from design, neuroscience, and computational biology, we examine how individuals learn to cooperate and form collective behavior through local, nonverbal action. In Tä, participants sit around a table in front of several piles of pebbles and follow simple rules to manipulate them. Without speaking or directly communicating, participants begin by moving pebbles in their own way, but over time, their actions synchronize, and give rise to stable yet evolving collective patterns. This process offers an experimental model for studying how group dynamics emerge in real-world settings. One of the unique features of our experiment is the use of an evolving participatory artwork as the experimental environment.. Although Tä is intentionally designed, its open-ended structure captures key features of natural social interaction: distributed decision-making, playful engagement, adaptation, and the spontaneous emergence of coordination. This allows us to investigate collective dynamics in a setting that is at once controlled, immersive, and ecologically rich. In this first presentation of the project , we describe our ongoing analyses of synchronization in participants' movements, as well as the formation, motion, and mixing of pebble groupings over time. We examine which interaction patterns arise reliably across performances, which depend on the composition and behavior of participants, and which may reflect more universal principles of collective organization. In addition, we relate these emergent dynamics to qualitative measures of human behavior and interaction, with the goal of clarifying how subjective experience, observable behavior, and group-level organization become linked during participatory social art performance.

KEYWORDS
self organization, synchrnoization, learning, emergant group formation, complex systems

HFSP Reference Number: RGP009/2025
HFSP Award Category: Research Grant
HFSP Award Year: 2025

MULTIPLEXED BIOMATERIAL TISSUE PERTURBATIONS

John Hickey
Duke University, USA

ABSTRACT
We developed a method for multiplexing biomaterial perturbations in vivo and reading out the local cellular environment with spatial-omics. Specifically we made microparticle hydrogels and injected into mice that were differentially barcoded. We then harvested the tissues in which the particles were injected and then sectioned and imaged using CODEX multiplexed imaging using a Phenocycler Fusion instrument with 50 antibodies targeting 21 cell types. We then developed and used computational algorithms including machine learning and bioinformatic algorithms to realize which cells were enriched around biomaterials and which multicellular neighborhoods were associated. This platform represents a unique way in which we can screen a large number of perturbations in vivo and associate with multicellular responses, representing an important avenue to test biomaterials, create many more perturbations, develop and test new therapies, and link for the first time associations with large scale cellular responses in tissues.

KEYWORDS
Spatial-omics, Biomaterials, Multiplexed screening, Cellular organization, Tissue biology, Computational multicellular analysis

HFSP Reference Number: RGEC26/2025
HFSP Award Category: Research Grant Early Career
HFSP Award Year: 2025

UNCOVERING THE COMPLEXITIES OF INTRINSIC AND RNA-DEPENDENT PHASE BEHAVIORS OF FUSED IN SARCOMA

Yuri Hong
Max Planck Institute of Molecular Cell Biology and Genetics (MPI-CBG), Germany

Vicky Liu[2]; Mrityunjoy Kar[3]; Min Kyung Shinn[2]; Laura T. Vogel[4]; Claus A. M. Seidel[4]; Anthony A. Hyman[1]; Rohit V. Pappu[2]
[1]Max Planck Institute of Molecular Cell Biology and Genetics; [2]Washington University; [3]Leibniz Institute of Polymer Research Dresden; [4]Heinrich Heine University Düsseldorf

ABSTRACT
Proteins containing RNA recognition motifs (RRMs) and intrinsically disordered regions (IDRs) play central roles in driving biomolecular condensate formation. Recent work has revealed important distinctions among RRM–IDR proteins, governed by factors including the sequence-encoded inter-domain interactions. For proteins such as Fused in Sarcoma (FUS), extant studies have focused almost exclusively on macrophase separation, known as liquid-liquid phase separation, which gives rise to micron-scale condensates. However, recent discoveries of significant clustering below the saturation concentration have proven confounding for a purely macrophase separation centric view. Furthermore, although FUS interacts with specific nuclear regulatory RNAs, the impacts of cognate versus non-cognate RNAs on FUS phase behavior remains enigmatic. We used a combination of computational approaches and in vitro biophysical measurements to uncover the complex hierarchy of processes that combine percolation, microphase separation within percolated networks, and macrophase separation aided by clustering of microphases. These findings identify a hierarchy of processes and species that are governed by the FUS-specific pattern of inter-domain attractions and repulsions. We studied the effects of cognate and non-cognate RNA molecules on the hierarchical phase behavior of FUS. For strongly binding, cognate RNAs, we observe a duality of effects. At low RNA-to-protein ratios, the cognate RNAs enhance the driving forces for percolation by increasing the effective valence, whereas this enhancement is reversed at higher RNA-to-protein ratios. The effects RNAs on FUS phase behaviors are sequence- and length-dependent. Strikingly, above FUS concentrations of ~250 nM, the influence of different RNA molecules converges, such that different RNAs provide a generic enhancement of network connectivity and mesoscale assembly. Taken together, our findings demonstrate that the intrinsic phase behaviors of FUS show hierarchical, concentration-dependent complexities that are glossed over in simple, LLPS-centric models. Additionally, RNA molecules act tunable regulators of FUS phase behavior, with their sequence-specific versus generic effects being influenced by the absolute concentration of FUS.

KEYWORDS
FUS, Microphase separation, Percolation, Macrophase separation, Biomolecular condensates

HFSP Reference Number: CDF0031/2025
HFSP Award Category: Cross Disciplinary Fellowship
HFSP Award Year: 2025

BEYOND BLUE: SPECTRAL PURITY, POLARIZATION, AND TIMING SHAPE INTERACTIONS OF MORPHO BUTTERFLIES WITH SYNTHETIC COLORS

Marko Ilić
University of Ljubljana, Slovenia

Uroš Cerkvenik[1]; Florian Muijres[2]; Matthieu Gallant Canguilhem[3]; Violaine Llaurens[4]; Vincent Debat[4]; Gregor Belušič[1]
[1]University of Ljubljana; [2]Wageningen University; [3]College de France; [4]Muséum National d'Histoire Naturelle (MNHN)

ABSTRACT
Morpho butterflies are famous for their vivid blue wings, and patrolling males readily investigate blue objects—behaviour consistent with detecting and evaluating conspecifics. But "blue" in nature is not a single wavelength: forest illumination shifts throughout the day as well as with shade and sun flecks, and wing reflections can vary in both spectral composition and polarization. To identify which visual cues trigger approach behaviour, we need stimuli whose spectrum, brightness, and timing can be controlled precisely while still being presented in a large, naturalistic flight space. Here we introduce LightCube, a system designed to test how butterflies respond to defined features of a visual signal, namely the spectrum, intensity, flashing frequency, and polarization. LightCube uses 16 monochromatic LEDs that together with a white integrating sphere and a switchable polarizing filter produce a spatially uniform, glare-free stimulus with a large angular extent and arbitrary spectral composition. LightCube can closely reproduce a chosen target spectrum and allows for independent control of overall intensity and temporal pattern. The temporal profile is fully programmable, allowing steady presentations or defined on/off and dynamic sequences. We applied the LightCube to quantify attraction in free-flying Morpho helenor in a large butterfly pavilion at Burgers' Zoo, Arnhem, The Netherlands. We defined attraction as inspection flights, scored as obvious deviations from the butterfly's ongoing flight path toward the stimulus. Behaviour was recorded with a camera setup, allowing objective counting of inspection visits and quantification of inspection behaviour in 3D. In addition to controlled-enclosure experiments, LightCube has been used in the rainforest of French Guiana to present calibrated stimuli under natural forest lighting, making it possible to link light cues to behaviour measured in the ecological context where these signals evolved. We found that polarization increased inspection numbers, indicating that Morpho attraction is influenced by more than spectral composition alone. Additionally, butterflies showed stronger attraction when the stimulus contained a purer blue component. Together, these results suggest that Morpho approach behaviour integrates multiple visual modes —spectral composition and polarization—potentially improving detection and evaluation of biologically relevant signals under complex lighting conditions. LightCube enables precisely controlled tests of visual preferences across species and ecological contexts.

KEYWORDS
Morpho, compound eyes, visual ecology, flight behaviour, attraction

HFSP Reference Number: RGP005/2023
HFSP Award Category: Research Grant Program
HFSP Award Year: 2023

MIRROR IMAGE FLOWERS-BREAKING BILATERAL SYMMETRY AND CONSERVATION GENETICS BOUNDARIES

Nicola Illing
University of Cape Town, South Africa

Spencer Barrett[1]; Eva Deinum[2]; Michael Lenhard[3]
[1]Toronto University; [2]Wagenignen University; [3]Potsdam University

ABSTRACT
Breaking bilateral symmetry is rare in both animals and plants. In mirror-image flowers, symmetry is broken by the reproductive organs being asymmetrically positioned, with the style deflected either to the left or the right of centre - a phenomenon known as enantiostyly. Although plants bearing both left- and right-handed flowers on the same individual are relatively common, evolutionary fixation of stylar handedness at the individual level is rare. Populations composed of left- and right-morphs are currently known only in Wachendorfia, Barberetta and Cubanicula (all in the Haemodoraceae) and Heteranthera multiflora. Our study investigates the genetic, cellular, evolutionary and ecological basis of this phenomenon. We show that the direction of style and stamen displacement in Wachendorfia and Barberetta is controlled by a hemizygous supergene present only in right-morph individuals. The supergene contains two key components: a functional microRNA (miR156), expressed in styles, and an auxin biosynthetic gene (YUC-R), expressed in stamens. Naturally occurring mutants lacking miR156 or YUC-R display misdirected styles or stamens, respectively, confirming that these genes overwrite the default left-handed floral state. The current hypothesis is that enantiostyly evolved to ensure efficient cross-pollination between the reciprocal stamens and styles of left- and right-handed flowers. Genetic paternity tools developed to test pollen exchange between left- and right-morph flowers have been extended to conservation genetics research, enabling the assessment of genetic diversity in the endangered Clanwilliam Cedar, Widdringtonia cedarbergensis, and informing its replanting programme.

KEYWORDS
dimorphic enantiostyly, supergene, conservation genetics, Clanwilliam cedar Widdringtonia cedarbergensis

HFSP Reference Number: RGP0036/2021
HFSP Award Category: Research Grant
HFSP Award Year: 2021

STATISTICAL ANALYSIS OF SMALL-INTEGER RATIOS IN BIOACOUSTICS AND MUSIC

Yannick Jadoul
Sapienza University of Rome, Italy

Tommaso Tufarelli[1]; Chloé Coissac[1]; Marco Gamba[2]; Andrea Ravignani[1]
[1]Sapienza University of Rome; [2]University of Turin

ABSTRACT
Rhythm is a fundamental aspect of human social interactions, from coordination of everyday tasks to joint musical performance. Rhythmic structure is ubiquitous in human culture as well as in nature, but is hard to capture in all its complexity. One common pattern in human music are temporal intervals whose relative durations can be expressed as small-integer ratios. For example, the durations of a quarter note and an eight note are related in a 2:1 ratio (Roeske et al., 2020). Recent work has found that the small-integer ratio categories do not just occur in most human musical cultures, but also in a broad range of animal species' vocalizations or behavioral displays. However, biological systems are noisy, and empirically measured intervals rarely form an exact small-integer ratio, and so, statistical methods are necessary to objectively assess whether an observed behavioral intervals approximately conform to a specific integer ratio. We explain a commonly-used approach for assessing the presence of integer ratio categories in temporal sequences, and then mathematically assess whether this leading integer ratio analysis method in behavioral research makes valid statistical and biological assumptions. In particular, we (1) make the temporal properties of empirical ratios explicit, both in general and for the typical use in the literature; (2) show how the choice of ratio formula affects the probability distribution of rhythm ratios and ensuing statistical results; (3) provide guidance on how to carefully consider the assumptions and null hypotheses of the statistical analysis; (4) present a comprehensive methodology to statistically test integer ratios for any null hypothesis of choice. Our observations have implications for both past and future research in music cognition and animal behavior: They suggest how to interpret past findings and provide tools to choose the correct null hypotheses in future empirical work.

KEYWORDS
rhythm, vocalization, social coordination, comparative approach, statistical assumptions

HFSP Reference Number: RGP0019/2022
HFSP Award Category: Research Grant
HFSP Award Year: 2022

INVESTIGATING INTERMITTENT BUZZING IN BUZZ-POLLINATION VIA BEE-SCALE MICRO-ROBOTIC SHAKERS AND POLLEN RELEASE STUDIES

Noah Jafferis
University of Massachusetts Lowell, USA

Charlie Woodrow[1]; Yuchen Kang[2]; Xiaowen (Edward) Chen[2]; Jennifer Shum[2]; Mario Vallejo-Marín[1]
[1]Uppsala University; [2]University of Massachusetts Lowell

ABSTRACT
Buzz-pollination, in which a pollinator generates vibrations to rapidly extract pollen from tube-like anthers, is used by approximately half of all bee species and over 20,000 species of plants, including crops such as tomato, kiwifruit, and blueberries. Our project investigates bee vibrations and bee-flower interactions during buzz-pollination via high-speed videography of real bees, custom bee-scale micro-robotic shakers ("robobuzzers"), and pollen release measurements and simulations. Using "robobuzzers" in addition to real bees allows us to vary design and operational parameters controllably (which is difficult or impossible to achieve with living bees), as well as to operate in regions of parameter space beyond what bees can achieve naturally, in order to better understand bee behavior and how 'optimal' it is.
Here we present some of our recent work and preliminary investigations into several open questions, such as: Does the temporal variation observed in bee buzzing amplitude have a benefit? And if so, what is the physical causation? Does vibration frequency effect pollen release separately from acceleration? Through experiments using "robobuzzers" to elicit pollen release from five plant species, we found that intermittent vibrations resulted in 60-70% more pollen release than continuous vibration of the same total vibration duration, which provides at least one explanation of why bees utilize similar behavior. While we obtained similar results with a large mechanical shaker, we found that the "robobuzzers" released approximately half of the amount of pollen as the shaker did. This is likely due to the differences in coupling and mass – for example, the external shaker was connected directly between mechanical ground and the anther, while the "robobuzzers" were mounted on the anther with their other side freely vibrating, which is potentially a closer match with actual bee behavior. Experiments are ongoing to further investigate such effects of shaker-anther coupling and shaker mass on pollen release, particularly as a function of vibration frequency. We will also describe our preliminary pollen-release simulations and updates on "robobuzzer" design with articulated heads and improved modularity for multi-parameter studies.

KEYWORDS
Bees, Biomechanics, Buzz-Pollination, Micro-Robotics

HFSP Reference Number: RGP0043/2022
HFSP Award Category: Research Grant
HFSP Award Year: 2022

MORPHOLOGICAL BASES OF BLUE IRIDESCENCE DIVERSITY ACROSS MORPHO BUTTERFLIES

Mariana Jaramillo
University of Antioquia, Colombia

Marco Giraldo[1]; Juliana Sosa[1]; Ana Valencia[1]; Melanny Silva[1]; Vincent Debat[2]; Gregor Belusic[3]
[1]University of Antioquia; [2]Muséum National d'Histoire Naturelle (MNHN); [3]University of Ljubljana

ABSTRACT
Morpho butterflies are renowned for their brilliant structural coloration, classically attributed to the "Christmas tree" ridge architecture of their dorsal wing scales. However, across the genus, wing coloration varies markedly in hue, intensity, angular distribution, and polarization properties. This diversity raises the question of whether variation of the canonical structural model adequately explains the optical variability observed among Morpho species. To examine how nanostructural variation shapes optical output, we studied the thirty species of the Morpho genus, by combining spectrophotometry, scatterometry, polarization-resolved reflectance measurements, scanning and transmission electron microscopy (SEM and TEM), and finite-difference time-domain (FDTD) simulations based on experimentally-derived geometries. Morphological analyses reveal substantial interspecific differences in ridge spacing, lamellar thickness, multilayer organization, ridge inclination, and degree of structural disorder. In several species, the scale architecture departs from the classical Christmas tree configuration. These structural differences correlate with pronounced variation in spectral reflectance and polarization signatures. Although all species exhibit coloration arising from multilayer interference within the ridges, the strength and spectral distribution of polarization-dependent reflectance vary considerably. FDTD simulations reproduce the principal spectral and polarization features and show that small changes in nanostructural parameters, particularly lamellar periodicity, ridge separation and thickness, can generate distinct optical responses.
Lastly, the reflectance of the wings strongly depends on the spectral properties of the natural illuminant, e.g. morning skylight, daylight, or light in the green understory. We show extensive variation across species in the coloration mechanisms that create robust patterns of reflectance, i.e. that are largely independent of the illuminant type, or patterns that change with the illuminant, which might affect camouflage or intraspecific recognition. Overall, our results indicate that Morpho wing coloration is not based on a single shared architectural organization, but on a structurally diverse panel of traits in which subtle morphological variations produce species-specific spectral and polarization outcomes.

KEYWORDS
butterfly coloration, multilayers, FDTD, iridescence, polarization

HFSP Reference Number: RGP005/2023
HFSP Award Category: Research Grant
HFSP Award Year: 2023

TRANSLATION REWIRING DURING EMBRYONIC DIAPAUSE

Ida Marie Astad Jentoft
Research Institute for Molecular Pathology (IMP), Austria

Sascha Amann; Irina Grishkovskaya; Manuel Matzinger; Karl Mechtler; David Haselbach; Andrea Pauli
Research Institute of Molecular Pathology (IMP)

ABSTRACT
Animal life has evolved diverse strategies to survive and reproduce during periods of environmental stress. A prominent example is embryonic diapause—a reversible, dormant state in which embryonic development is paused until favorable conditions return. In the annual killifish, Nothobranchius furzeri, diapause can persist for months or even years, serving as a critical mechanism to survive the periodic desiccation of its natural habitat. To sustain long-term dormancy, diapause is characterized by a profound reduction in protein synthesis. Through global proteome profiling comparing dormant and developing killifish embryos, we identified the upregulation of conserved vertebrate ribosome dormancy factors. Using cryo-electron microscopy and mass spectrometry of isolated ribosomes, we demonstrate that these factors bind to ribosomes during diapause, directly contributing to the global shutdown of protein synthesis. Single-embryo proteomics of pre-diapause embryos revealed heterogeneous expression of these dormancy factors, reflecting the stochastic nature of diapause entry. Similarly, proteomic profiling during diapause exit showed a rapid depletion of these factors once development resumes, which together suggest that ribosome inhibition may actively drive the establishment of the dormant state. To determine whether inducing ribosome dormancy in early embryos is sufficient to promote diapause, we are utilizing a newly established photo-caged mRNA technique for the timed over-expression of dormancy factors. Furthermore, ongoing genetic perturbation experiments assess the necessity of translational repression for successful long-term survival during diapause. Together, our data reveal conserved mechanisms of translational arrest during embryonic diapause. These findings highlight the potential of the killifish model for discovering universal regulators of dormancy, thereby advancing our understanding of the fundamental principles underlying cellular and organismal suspended animation.

KEYWORDS
Diapause, dormancy, translation, ribosome hibernation

HFSP Reference Number: LT0029/2025-L1
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2025

TRACING THE EVOLUTIONARY ORIGINS OF RNA-BINDING PROTEIN NETWORKS: GIARDIA AS A MODEL FOR BASAL EUKARYOTIC INNOVATION

Aaron Jex
WEHI, Australia

Balu Balan
WEHI

ABSTRACT
RNA-binding proteins (RBPs) are fundamental post-transcriptional regulators in eukaryotes, orchestrating RNA splicing, transport, degradation, translation, and translational repression. Remarkably, the eukaryotic RBPome is largely conserved from yeast to humans, yet many of these proteins are absent or rudimentary in prokaryotes, suggesting that "eukaryotic-innovative" RBPs emerged early in eukaryogenesis. However, when and in what biological context these critical regulatory systems arose remains unclear. We search for insights into the earliest detectable origins of eukaryotic RBP networks using Giardia duodenalis, as a member of a basal eukaryotic lineage that diverged from the eukaryotic trunk approximately one billion years before the emergence of yeast. Our research program integrates phylogenomic, molecular, and functional approaches to characterise RBP evolution using Giardia as a minimal model system. We constructed a comprehensive phylogenomic atlas of RBP families across the tree of life, revealing that the eukaryotic RBPome was shaped by ancestral bacterial and archaeal systems alongside the emergence and expansion of novel eukaryotic RBP families. Domain topology analysis, domain co-occurrence mapping, and intrinsically disordered region (IDR) profiling demonstrate that Giardia RBPs exhibit simplified modular architectures that subsequently expanded in later eukaryotes, providing insight into the evolutionary trajectory of RBP complexity. To define the Giardia RBPome, we integrated domain- and structure-informed annotation with transcriptomics, proteomics, and PAR-CLIP-based RNA–protein interactome capture. This multi-omics approach revealed a diverse repertoire of both canonical RBPs and non-canonical "moonlighting" RBPs—metabolic enzymes and structural proteins that also bind RNA. We selected representative eukaryotic-innovative RBPs for detailed functional characterisation, including early Pumilio homologs (PUF and PUM), DEAD-box helicases (DDX3 and EIF4A), and the moonlighting factor phosphoglycerate kinase (PGK). Using targeted genetic knockouts via CRISPRi and enhanced CLIP-seq, we mapped direct RNA targets and regulatory networks for each RBP, revealing conserved regulatory phenotypes and RNA-binding specificities consistent with their eukaryotic counterparts. Strikingly, we demonstrated that recombinant versions of these Giardia RBPs undergo liquid-liquid phase separation (LLPS) both in vitro and in cellular contexts, with and without RNA. This condensate-forming behaviour—a hallmark of modern eukaryotic RBPs involved in membraneless organelle assembly—indicates that the biophysical principles underlying RBP-mediated compartmentalisation emerged early in eukaryotic evolution.

Our findings position Giardia as a tractable experimental model for studying the origins of eukaryotic post-transcriptional control. The presence of simplified yet functional RBP architectures, conserved RNA regulatory networks, and phase-separation capabilities in this basal eukaryote suggests that sophisticated RBP regulation was established at the dawn of eukaryogenesis. These early innovations likely played a pivotal role in eukaryotic emergence and diversification, enabling the complex gene expression programs that distinguish eukaryotes from prokaryotes. By characterising RBP evolution in Giardia, we provide fundamental insights into how ancient molecular innovations gave rise to the regulatory complexity that defines modern eukaryotic life.

KEYWORDS
RNA-binding proteins, eukaryotic evolution, LECA

HFSP Reference Number: RGY0075/2017
HFSP Award Category: Research Grant
HFSP Award Year: 2017

UNVEILING HISTORICAL HYDRODYNAMIC CUES AS A SENSORY LANDSCAPE FOR AQUATIC NAVIGATION TASKS

Gurvan JODIN
École Normale Supérieure de Rennes (ENS Rennes), France

James Herbert-Read[1]; Dixia Fan[2]; Nadia Maria Hamilton[1]; Bastien Lagneaux[3]; Ang Li[2]; Aoming Liang[2]; Hannah Mac Gregor[1]; Thomas Omarini[3]; Corentin Porcon[3]; Florence Razan[3]; Boai Sun[2]; Siming Wang[1]; Yi Zhu[2]
[1]Cambridge University; [2]Westlake University [3]ENS Rennes, CNRS

ABSTRACT
Animals moving in water leave behind complex hydrodynamic disturbances—coherent pressure, velocity, and vorticity structures that persist for seconds and encode information about the identity, size, posture, and movements of their generators. This project investigates how fishes detect, interpret, and use these "hydrodynamic fingerprints" to guide search, following, and avoidance behaviours in environments where visual cues are limited. Combining behavioural ecology, sensory biology, fluid mechanics, electronics engineering, and artificial intelligence, we develop an integrated experimental–computational framework to reveal the mechanisms by which aquatic animals exploit physical traces of past events. We constructed custom experimental arenas—a Y-maze for decision-making tasks and an open maze for free exploration—equipped with high-fidelity arrays of underwater differential pressure sensors. These MEMS-based sensors achieve sub-Pascal sensitivity (~0.15–0.2 Pa), enabling direct measurement of the weak, transient pressure fields generated by freely swimming fish. A distributed wireless acquisition network synchronises up to 100 pressure sensing points with infrared videography, ensuring precise temporal alignment between hydrodynamic disturbances and animal movements. To capture the full spatial structure of wakes, we developed a digital twin of the experiments using low-fidelity computational fluid dynamics (Boundary Data Immersion Method), validated against empirical pressure recordings and dye-visualised vortices. Across the first two years of the project, we quantified the scale, structure, and lifetime of hydrodynamic cues. Vortex signatures typically persisted seconds, and pressure minima due to fish motions scaled predictably with swimming speed and distance. Using AI tools—YOLO-based tracking, autoencoders, and recurrent neural networks—we are mapping fish posture and kinematics to the hydrodynamic cues they produced. Behavioural tests demonstrated that fish are significantly more likely to follow conspecifics when entering a decision point shortly (<10s) after them, even in darkness and turbidity, indicating use of non-visual hydrodynamic information. The project now advances towards mapping encountered cues to the movement decisions of other fish. To go further, we are engineering aquatic actuators capable of either disrupting real wakes or generating artificial ones that mimic predator or prey signatures. These experiments will provide the first direct tests of whether historical hydrodynamic cues alone can trigger following, tracking, or avoidance responses in real animals.

KEYWORDS
fish navigation, hydrodynamic, sensor arrays, computational fluid dynamics, computer vision

HFSP Reference Number: RGY0059/2022
HFSP Award Category: Research Grant Early Career
HFSP Award Year: 2022

SYSTEM-WIDE FLOW DYNAMICS IN DEVELOPING BIOLOGICAL NETWORKS

Eleni Katifori
University of Pennsylvania, USA

Alex Mayer[2]; Sidney Holden[2]; Laureline Julien[3]; Xinbo Li[1]; Aaron Winn[1]; Daisuke Noto[1]; Elizabeth Jones[4]; Annemiek Jm Cornelissen[3]
[1]University of Pennsylvania; [2]Flatiron Institute; [3]CNRS & Paris Cité University; [4]KU Leuven

ABSTRACT
Flow networks in animals—such as the arterial, venous, and lymphatic systems—are widely believed to emerge through self-organization. Mechanical stresses are thought to play a central role in this developmental process. However, these mechanical signals are not static; they are highly dynamic in space and time. What role do these dynamics play in shaping vascular development? With support from our HFSP award, we investigate flow systems in two evolutionarily distant organisms: the avian yolk sac as a model of vertebrate vasculature, and the gastrovascular canal system of the jellyfish. To understand the role of mechanical dynamics, we require flow measurements not only locally, but across the entire organ or organism. In this talk, we present our experimental and theoretical approaches to acquiring and interpreting such whole-system flow data. In the jellyfish, we use particle image velocimetry (PIV) to quantify flow within the canals and develop a theoretical model that incorporates whole-body deformations to predict flow throughout the animal as the animal is deforming while swimming. In the avian yolk sac, we combine kymograph analysis with homogenization methods to reconstruct flow dynamics across the entire vascular network. With comprehensive flow maps in both systems, we present recent results showing how whole-organism flow measurements reveal mechanisms of flow facilitation and redistribution that are not apparent from local data alone. These findings allow us to directly link dynamic flow patterns to mechanical stresses and their role in shaping developmental processes.

KEYWORDS
vasculature, mechanics, flow, dynamics, development, complex networks

HFSP Reference Number: RGP015/2023
HFSP Award Category: Research Grant
HFSP Award Year: 2023

VISUAL PATHWAYS THROUGH THE ANTERIOR OPTIC TUBERCLE IN THE PAPILIO BUTTERFLY BRAIN

Michiyo Kinoshita
The Graduate University for Advanced Studies, Japan

Setfanni Jahn[2]; Naomi Takahashi[1]; Jenny Y. Sung[1]; Agnish Prusty[1]; Mieko Kono[1]; Uwe Homberg[2]
[1]The Graduate University for Advanced Studies; [2]Marburg University

ABSTRACT
The Japanese swallowtail butterfly, Papilio xuthus, detects flowers using multiple visual cues: color, brightness, and polarization. These visual cues are processed separately and in parallel within the optic lobe, the primary visual processing center, and are subsequently conveyed to distinct regions of the central brain for integration and motor command generation. The anterior optic tubercle (AOTU), located in the central brain and connected to the optic lobe via a massive fiber tract, is a strong candidate for integrating these visual cues. The AOTU consists of upper and lower units. While the lower unit has been intensively studied as a relay station for sky compass navigation in many insects, the function of the upper unit (AOTU-UU) remains poorly understood. To determine the neural pathways through the AOTU-UU, we first reconstructed the neuropils of the central brain through synapsin immunostaining of whole-mount brains. We then mapped the neural tracts connecting the AOTU-UU with the optic lobe and other regions of the central brain by injecting a neuronal tracer into the AOTU-UU and analyzing the labeling patterns. We identified all neuropiles of the central brain based on a Drosophila brain template published in 2014. The AOTU-UU is connected to the optic lobe via the anterior optic tract, which contains numerous fine fibers originating from at least three cell types. In contrast, output neurons from the AOTU-UU project broadly to multiple regions of the central brain including the lateral accessory lobe, a locomotion center, and the gnathal ganglia, which are gustatory sensory centers. These identified neural networks provide an anatomical basis for further physiological studies and suggest that the AOTU-UU serves as a high-order processing area for visual information.

KEYWORDS
Vision, Brain, Neural circuit, Lepidoptera

HFSP Reference Number: RGP023/2023
HFSP Award Category: Research Grant
HFSP Award Year: 2023

THE MECHANICAL PROPERTIES OF LEAVES AS SANDWICH STRUCTURES

Eva Knight
Imperial College London, UK

John Dear Freng[1]; Yuancheng Yang[2]; Louis Youssef[3]; Nancy Walker[4]; Lamha Kumar[3]; Haibao Liu[2]; Anja Geitmann[3]
[1]Imperial College London; [2]Queen Mary University of London; [3]McGill University; [4]Yale University

ABSTRACT
The functioning of plant leaves must reconcile the optimization of structural features for mechanical stability with the needs for optimal photosynthesis. The inner photosynthetic tissues constitute the core of the sandwich-like leaf structure. Due to its presumed soft properties, the mechanical role of the mesophyll has not been explored. Here, uniaxial tension and vibration bending experiments have been conducted on rectangular sections of leaf material to obtain the leaves overall tensile and bending moduli. The leaf material was extracted from freshly grown leaves of Arabidopsis of a wild type and of a mutant type (CGR30X) which has larger mesophyll air spaces. Structural data of these leaves were obtained using X-ray imaging. Moreover, at the cellular level, mechanical properties of cell walls were tested using Atomic Force Microscopy (AFM). The mechanical data combined with the structural data were used to compare mesophyll and epidermis. The mechanical response of the two Arabidopsis strains were assessed to investigate how the mesophyll porosity contributes to the mechanical stability of the whole leaf.

KEYWORDS
mesophyll, epidermis, biomechanics, sandwich structure, Young's modulus

HFSP Reference Number: RGP010/2023
HFSP Award Category: Research Grant Program
HFSP Award Year: 2023

ELECTROSTATIC MATRIX ASSEMBLY IN REGENERATION: A COLLAGEN-SILK FIBROIN JELLYFISH BIOMIMETIC PLATFORM

Vladislav Koren
Weizmann Institute of Science, Israel

Carl Modes[2]; Chiara Sinigaglia[3]; Ulyana Shimanovich[1]
[1]Weizmann Institute of Science; [2]Max Planck Institute of Molecular Cell Biology and Genetics; [3] Sorbonne University

ABSTRACT
Jellyfish exhibit extraordinary regenerative capacity, with the ability to restore up to half of their body mass within relatively short timeframes following severe injury or fragmentation. While the majority of jellyfish biomass consists of collagen, a structural protein widely used in tissue engineering and wound-healing applications, the presence of collagen alone cannot sufficiently explain such rapid and extensive regeneration. Increasing evidence suggests that negatively charged glycosaminoglycan (GAG)-like polysaccharides within the extracellular matrix, and their electrostatic and structural interactions with collagen, may play a central role in modulating tissue remodeling, cell signaling, and matrix reorganization during regeneration. Silk fibroin, a biocompatible protein extensively applied in regenerative medicine, presents a useful analogue to these GAG-like components. Although structurally distinct from polysaccharides, silk fibroin possesses an overall negative charge under physiological conditions and exhibits highly tunable physicochemical properties, enabling the formation of matrices that mimic diverse tissue microenvironments without eliciting significant immune responses. Its capacity to interact with collagen through electrostatic forces, hydrogen bonding, and hierarchical assembly makes it a promising model system for probing matrix-driven regenerative mechanisms. By systematically investigating collagen-silk fibroin interactions as a simplified biomimetic platform, this work aims to elucidate how charge mediated matrix organization, composite assembly and multiscale structural hierarchy may contribute to large-scale tissue regeneration. Understanding these fundamental interactions could provide insight into the regenerative strategies of jellyfish and inform the rational design of advanced biomaterials for wound healing and tissue engineering applications.

KEYWORDS
extracellular matrix, silk protein, regeneration, jellyfish, collagen

HFSP Reference Number: RGP020/2024
HFSP Award Category: Research Grant
HFSP Award Year: 2024

CELL FATE MEMORY IS HERITABLE BUT TRANSIENT AND REVERSIBLE

Nitu Kumari
Northwestern University Feinberg School of Medicine, USA

Hanxiao Sun; Yogesh Goyal
Northwestern University Feinberg School of Medicine

ABSTRACT
Cell fate determination is fundamental to how organisms develop and adapt, yet how genetically identical cells adopt divergent fates under the same external stimulus remains a central open question. Cancer cells offer a powerful model to study this problem: under drug treatment, rare cells within a genetically uniform population survive and proliferate while most other cells are eliminated. Understanding what governs this fate dichotomy and whether it is remembered across cell divisions has profound implications not only for cancer therapy but for our broader understanding of cell fate commitment and maintenance. We developed an integrative framework combining DNA barcoding with single-cell multi-omics to trace how individual cells within isogenic KRAS-mutant cancer populations adopt resistant or sensitive fates across diverse drugs. By connecting each cell's molecular identity to its ultimate fate, we revisited a foundational question first posed by Lederberg in the 1950s: are resistant cells pre-existing within the population, or do they arise de novo in response to treatment? We demonstrate that a rare subset of cells reproducibly survived both targeted and chemotherapies, and that their propensity to resist was already encoded in their transcriptional, morphological, and chromatin accessibility profiles before any drug exposure. Targeted therapies triggered far greater molecular reprogramming in surviving cells than chemotherapy, and critically, these changes were fully reversible upon drug removal, establishing non-genetic mechanisms as the primary determinants of fate. If resistance is non-genetically encoded, a key prediction follows: fate memory should be transmitted to daughter cells but not persist indefinitely. To test this directly, we tracked clonal lineages through successive generations before drug challenge. Specific clones reproducibly survived, demonstrating that resistance propensity is heritable over short generational timescales of four to five cell divisions. Beyond this window, memory progressively faded, and previously sensitive lineages newly acquired resistance. We conclude that cell fate memory is written, inherited, and eventually erased, defining a temporal window during which a cell's resistant identity is maintained. What molecular machinery sustains this transient memory? We identified the AP-1 transcription factor complex as a central regulatory switch: inhibiting AP-1 during drug exposure significantly reduced resistance, while activating AP-1 prior to treatment rewired cells into a drug-tolerant state and enhanced survival. Perturbation of additional identified cell-state regulators similarly abolished resistance, demonstrating that specific molecular factors are required to both initiate and maintain the resistant fate.

Our work establishes that cell fate decisions in cancer-therapy resistant cells are governed by a transient, reversible cellular state with defined molecular regulators that can be functionally targeted, opening new avenues for controlling fate outcomes by manipulating the machinery of cell state memory.

KEYWORDS
cell state, cell fate, memory, barcode, heritability

HFSP Reference Number: LT0024/2025-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2025

DEVELOPMENT OF A SMART EXPERIMENTAL DEVICE FOR MEASURING AND MANIPULATING THE HYDRODYNAMIC STATE OF A TANK WITH SUBMERGED BODIES

Bastien Lagneaux
ENS Rennes, CNRS, France

Corentin Porcon; Thomas Omarini; Florence Razan; Gurvan Jodin
ENS Rennes, CNRS

ABSTRACT
Understanding how fish perceive and respond to water disturbances generated by nearby swimmers is central to explaining collective behavior, predator avoidance, and prey tracking. However, measuring these hydrodynamic signals typically requires invasive and/or computationally expensive techniques such as Particle Image Velocimetry (PIV), which limits experimental flexibility.
This work presents a novel, noninvasive framework that combines video recordings and pressure sensor data to reconstruct the hydrodynamic environment around freely swimming fish. Using artificial intelligence–based image segmentation, we automatically extract fish kinematics from laboratory videos and generate corresponding digital twin simulations using Computational Fluid Dynamics (CFD). Although these simulations are lower in physical fidelity than PIV, they provide spatially rich flow estimates with significantly reduced cost and computational demand.
To improve accuracy, we investigate data fusion strategies that integrate simulation outputs with distributed pressure sensor measurements. Initial reduced-order modeling approaches proved insufficient to fully capture the complex tank dynamics, motivating the exploration of modern machine learning methods, including transformer-based models, variational autoencoders, and diffusion-based generative techniques. A dedicated multi-source and multi-fidelity benchmark dataset is being constructed to systematically evaluate these approaches.
This framework aims to provide biologists with an accessible tool for studying how fish use hydrodynamic cues, enabling scalable and minimally invasive experiments. Beyond behavioral ecology, this approach may also inform the design of bio-inspired underwater robots that sense and respond to flow disturbances.

KEYWORDS
Hydrodynamic, Digital Twin, Noninvasive Flow Reconstruction, Automated Experiments, Data Fusion

HFSP Reference Number: RGY0059/2022
HFSP Award Category: Research Grant Program
HFSP Award Year: 2022

INVERSE LACTATION: LARVAL SECRETIONS CONTROL REPRODUCTIVE DIVISION OF LABOUR IN ANTS

Adria C. LeBoeuf
University of Cambridge, UK

Helder Hugo; Nadine Anders; Yuqi Reitsema-Wang; Jean-Jacques Rafanomezantsoa; Bas Teusink; Brian L. Fisher
University of Cambridge

ABSTRACT
Social fluids typically flow from adults to offspring, with milk being the iconic example. Here we describe the reverse: in the ponerine ant Platythyrea mocquerysi, larvae provision adults through a specialised tubercle, and access to this "inverse lactation" determines which adult can act as reproductive. Platythyrea colonies often lack morphological queens. Instead, reproductive division of labour emerges dynamically among totipotent workers. We asked how reproductive roles are established in this system. Combining controlled experiments manipulating larval access with automated behavioural tracking, proteomics and transcriptomics of larval secretions, and electron microscopy of internal morphology, we found that access to larval tubercle secretions enables reproductive activation: only individuals with access to larvae initiated egg laying. Those engaging more frequently in larval feeding laid eggs faster, lived longer, and exhibited more brood care. Both larvae and adults feed on prey, yet reproductives appear to shortcut their own metabolic costs by co-opting larval metabolism. These findings reveal larvae as gatekeepers of adult reproductive status and demonstrate a novel form of metabolic division of labour: rather than reproductives receiving processed food from helpers, they extract resources directly from developing brood, extending their own lifespan in the process.

KEYWORDS
social evolution, reproductive division of labour, metabolic division of labour, Ponerinae, caste plasticity

HFSP Reference Number: RGP0023/2022
HFSP Award Category: Research Grant
HFSP Award Year: 2022

A SWITCHABLE OPTOCHEMICAL RNA TOOL FOR SPATIAL CONTROL AND IMMUNOMODULATION OF LOCAL TRANSLATION IN MICROGLIA

Kathrin Leppek
University of Bonn, University Hospital Bonn, Germany

Vivian Lobo[1]; Elarbi Chatir[2]; Jakob Rupert[3]; Meng-Meng Fu[4]; Dragomir Milovanovic[3]; Daniel L.J. Broere[2];
[1]University of Bonn, University Hospital Bonn; [2]Utrecht University; [3]Charité – Universitätsmedizin Berlin; [4]University of California, Berkeley

ABSTRACT
Widespread control of gene expression through translation has emerged as a key level of spatiotemporal protein expression regulation. Internal ribosomal entry sites (IRESes) provide a prominent mechanism by which ribosomes can confer greater gene regulation by mediating direct ribosome recruitment [1] [2]. We recently presented a set of technologies in embryos and cells to study cellular IRES activity as a new guideline to understand IRES-mediated translation [3]. We apply these tools to our HFSP-funded project. Here, we focus on microglia, the resident immune cells of the brain, and the first line of defense against pathogens by surveillance and phagocytosis. Microglia contain dynamic PEripheral Microglial Processes (PeMPs) that retract upon pathological insult. Local mRNA translation in these protrusions likely contributes to these responses, but only little evidence exists for this in microglia. We study the functional role of localized translation in the microglial response. We employ targeted RNA delivery including optimized RNA packaging and optochemical activation of a bioactive RNA tool to achieve spatial and temporal control of translation. Delivery of RNA cargo into microglia in itself is challenging. We optimized delivery of different mRNA reporters as cargo into mouse microglia cell lines and primary cells and evaluated their translation efficiency by luciferase assays, microscopy and flow cytometry readouts. We selected 21 candidate mRNAs reported to localize to and function in PeMPs. We next tested the untranslated regions (UTRs) of these candidate mRNAs in reporter mRNAs. We assess the role of these UTRs in translation and subcellular localization in primary microglia using luciferase assays and single-molecule FISH (smFISH), respectively. We also identify the minimal regulatory structural RNA elements in these UTRs that are responsible for mRNA translation and localization. Our team is engineering a complementary RNA photoswitch we termed SMARTswitch (Specific Molecular Azobenzene RNA Target) that enables a light-induced reversible structural change of this tool to optochemically regulate mRNA UTR elements with the goal to modulate translation in microglia. This RNA photoswitch enables us to study the function and physiological relevance of localized translation in microglial responses in a gene-specific manner, reversible, and controlled by light. This work is promising for engineering of microglial immune responses in vivo to allow spatial optochemical gene regulation through targeting of regulatory structural RNA elements in microglia. Thus, such synthetic RNA-based tools, directly relevant for RNA therapeutics [4], have the potential to allow immunomodulation of microglia in health and disease.

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KEYWORDS
mRNA translation regulation, microglia, immunomodulation, optochemical RNA tool, local translation, brain immune cells

HFSP Reference Number: RGEC32/2023
HFSP Award Category: Research Grant Early Career
HFSP Award Year: 2023

RESTORING HOMEOSTATIC LEVELS OF A HISTONE VARIANT COUNTERS AGING-ASSOCIATED CELLULAR DECLINE

Anastasios Liakos
University Medical Center Göttingen, Germany

Vassiliki Varamogiani-Mamatsi; Nihan Koç; Argyris Papantonis
University Medical Center Göttingen

ABSTRACT
Beyond their structural role in chromatin organization as the core components of nucleosomes, histone proteins are key regulators of molecular processes such as replication, DNA repair, and transcription. Cellular aging, a complex but conserved phenomenon across species, is characterized by a depletion in the levels of various histone proteins, thereby affecting chromatin structure and function. We focused on H2A.Z, a replication-independent histone variant of canonical H2A enriched at cis-regulatory elements, including promoters and enhancers. H2A.Z is markedly depleted from different models of cellular senescence, as well as in aging tissues. We restored homeostatic levels of H2A.Z in near-senescent human cells, and found this to be sufficient for sustain growth primarily by countering the senescence-induced global transcriptional suppression. Our findings highlight an underappreciated role of H2A.Z in maintaining cellular homeostasis, and suggest that chromatin-based interventions may represent a promising strategy for decelerating age-associated cellular decline and late-life disease.

KEYWORDS
Cellular senescence, aging, chromatin, H2A.Z

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HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2024

EMERGENT COLLECTIVE BEHAVIOR IN MULTI-AGENT REINFORCEMENT LEARNING AND ANALYTICALLY TRACTABLE MODELS

Samuel Liebana
University College London, UK

Samuel Lozano Iglesias[2]; Nishil Patel[1]; Miguel Ruiz-Garcia[2]; Andrew Saxe[1]
[1]University College London (UCL); [2]Complutense University of Madrid

ABSTRACT
We study the emergence of collective behavior in systems of interacting reinforcement learning (RL) agents with heterogeneous reward structures and individual performance. Our work combines two complementary approaches. On the one hand, we perform RL simulations to explore how coordination arises in a shared environment. On the other hand, we develop theoretical frameworks that simplify the environment and extend the teacher-student setting to multi-agent systems, which allows us to derive analytical descriptions of the resulting dynamics. By comparing these two approaches, we aim to identify common mechanisms underlying collective behavior across different levels of model complexity. In particular, we consider agents with different abilities and reward structures that encode distinct interaction tendencies, and we analyze how these differences influence the dynamics at the group level. This combined computational and analytical perspective provides a controlled setting to investigate how individual learning rules give rise to collective organization.

KEYWORDS
collective intelligence, reinforcement learning, theoretical machine learning

HFSP Reference Number: RGEC33/2024
HFSP Award Category: Research Grant Early Career
HFSP Award Year: 2024

CYCLICAL SEXUAL–ASEXUAL SELECTION DRIVES ADAPTIVE FUSION VIA REPRODUCTION TRADE-OFF

Gaowen Liu
Shenzhen Institutes of Advanced Technology, Chinese Academy of Sciences, China

ABSTRACT
Life-history trade-offs under fluctuating selection are central to ecology and evolution, yet we rarely observe—at molecular resolution—how transient adaptive states become genetically fixed. Here we challenge the fission yeast Schizosaccharomyces pombe to overcome a severe reproductive defect caused by loss of the critical fusion protein Prm1, using a high-throughput evolution regime of ~500 mitotic generations interleaved with 18 cycles of sexual differentiation. Across lineages, time-resolved transcriptomics reveals an early phase of oscillatory plasticity in mating and cell-cycle programs, while phenotypic tracking exposes a clear growth–fusion trade-off; integrating these layers shows that cell-cycle transcriptional programs co-vary with fluctuations in growth and fusion across cycles. In the most rescued lineage, stable adaptation is “locked in” when a 149-kb subtelomeric deletion becomes fixed, triggering subtelomeric position effects that silence the chromatin regulator clr5 and derepress a ste11-driven mating regulon. Genetic reconstruction maps a compensatory pathway through ste11, sms1, and the cell-surface effector adg2, which significantly rescues fusion despite the absence of Prm1. Finally, rational overexpression of adg2 achieves strong rescue with minimal growth penalty, indicating that the evolved trade-off reflects constraints of an accessible genomic route rather than an immutable biological limit. Together, our results connect fluctuating selection, genetic assimilation, and genome–epigenome–transcriptome rewiring in a tractable eukaryotic system.

KEYWORDS
evolution, meiosis, yeast, adaptive fusion

HFSP Reference Number: LT-001727/2017-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2017

EXPERIMENTAL MISSHAPES, MISTAKES AND MISFITS: SPOROMORPH MALFORMATIONS AND MASS EXTINCTIONS

Barry Lomax
University of Nottingahm, UK

Matthew Kentl[1]; Bas Van De Schootbrugge[2]; Cindy Looy[3]; Ivo Duijnstee[3]; Thijs Vandenbroucke[4]
[1]University of Nottingahm; [2]Utrecht University; [3]University of California Berkeley; [4]Ghent University

ABSTRACT
The sporomorphs (spore and pollen) fossil record is one of the richest records of past life available for palaeobiologists to study. This exceptional archive is a combination of productivity and preservation potential. As an example of this, Al Traverse suggested that an average hectare of woodland in eastern North America produces at least 3,000 litres of pollen per year, that a gram of siltstone can contain in excess of four million sporomorphs, and a single microscope slide can contain thousands of specimens. This unparalleled archive of past life allows for a detailed understanding of morphology and its biological variability. In turn this has led to the recognition that a number of mass extinctions and carbon cycle perturbations are evidenced by increases in the abundance of malformed sporomorphs. These malformations are postulated to have occurred through abiotic stress impacting on normal reproductive development, with UV-B radiation and volatile metals being identified as the primary drivers. Whilst the case for elevated UV-B and thus stratospheric ozone is well developed, work on the role of metals in driving malformations has received less attention. Using an experimental framework this presentation will discuss evidence for the role of metals in driving sporomorph malformations.

KEYWORDS
mass extinction, pollen, spores

HFSP Reference Number: RGP0066/2021
HFSP Award Category: Research Grant
HFSP Award Year: 2021

VARIATION IN SKULL MORPHOLOGY REFLECTS FUNCTIONAL SPECIALISATION IN REPTILIAN HEAD-FIRST BURROWERS

Anastasiia Maliuk
University College London, UK

Vinicius Anelli[2]; Morgane Taillades[3]; Anastasiia Sadetskaia[4]; Maxwell Olson[5]; Jungrui Hu[1]; Susan Evans[1]; Anthony Herrel[6]; Henrik Birkedal[4]; Dominique Adriaens[3]; Tiana Kohlsdorf[2]; Mehran Moazen[1]

[1]University College London; [2]University of São Paulo; [3]Ghent University; [4]Aarhus University; [5]Clark University; [6]Museum National d'Histoire Naturelle

ABSTRACT
Head-first burrowing is an extreme mode of subterranean locomotion that imposes high mechanical demands on the vertebrate skull. Amphisbaenians (Amphisbaenia), a clade of elongate, mostly limbless squamates, are among the most specialised head-first burrowers and show striking diversity in cranial architecture. Traditionally, amphisbaenians have been classified into a small number of cranial morphotypes (round, keel-shaped, spade-shaped and shovel-shaped), assumed to reflect distinct burrowing strategies (Gans, 1968). However, these categories remain largely qualitative, and it is unclear which aspects of cranial morphology are most strongly associated with functional specialisation versus phylogenetic history. The key question is whether functional specialisation is expressed through whole-skull divergence between morphotypes, or through modular, bone-specific changes constrained by phylogeny. Here, we quantify cranial variation in amphisbaenians using a CT-based dataset of 110 segmented skulls. Individual cranial bones and major cranial regions were segmented and labelled to enable volumetric and proportional comparisons across taxa. We analysed (i) relative cranial bone and regional volumes, and (ii) size-corrected linear proportions capturing skull geometry. Volumetric data were treated as compositional and analysed using centred log-ratio transformation, while proportional data were standardised prior to multivariate analysis. Principal component analyses (PCA) were performed separately for volumes and proportions, and visualised at multiple taxonomic levels, including within the family Amphisbaenidae and across all amphisbaenian families. Preliminary results indicate that relative cranial bone volumes overlap broadly across morphotypes, with variation expressed in a modular and bone-specific manner rather than as a single consistent shift across the skull. PCA based on cranial bone volumes captures a strong phylogenetic structure: family-level clustering is clearer than morphotype separation, and the most diverse family Amphisbaenidae forms a distinct region of morphospace with limited overlap with other families. In contrast, PCA based on size-corrected proportions shows a stronger morphotype-related signal within Amphisbaenidae, with major variation aligned along an axis of cranial robustness (relative skull width, height and cranial angle). Across all families, however, phylogeny remains the dominant structuring factor, reflecting the close association between morphotypes and deeper taxonomic divisions.

Together, these results suggest that cranial bone volumes are relatively conservative and primarily constrained by phylogenetic history, whereas proportional geometry may capture morphotype-related functional differentiation more clearly at shallower taxonomic levels. Ongoing work will extend this framework by applying landmark-based geometric morphometrics to individual cranial bones, enabling direct tests of which skeletal elements contribute most to functional specialisation in head-first burrowing reptiles.

Gans, C. 1968. Relative success of divergent pathways in amphisbaenian specialization. American Naturalist 102:34

KEYWORDS
Amphisbaenia, skull morphology, computed tomography (CT), geometric morphometrics.

HFSP Reference Number: RGP013/2025
HFSP Award Category: Research Grant
HFSP Award Year: 2025

PILI FOR NANOWIRES: SINGLE-CELL IMAGING OF BACTERIAL RESPIRATION VIA EXTRACELLULAR ELECTRON TRANSFER CORRELATED WITH GENETIC, BIOCHEMICAL, AND BIOPHYSICAL STUDIES IDENTIFY A NEW HYBRID TYPE 4 PILUS/TYPE 2 SECRETION SYSTEM FOR CYTOCHROME NANOWIRES

Nikhil Malvankar
Yale University, USA

Olivera Francetic[1]; Lisa Craig[2]; Carlos Salgueiro[3]
[1]Institut Pasteur; [2]Simon Fraser University; [3]NOVA University of Lisbon

ABSTRACT
Diverse anaerobic bacterial and archaeal species residing all over the planet respire without soluble electron acceptors using extracellular electron transfer (EET) to metals, electrodes of fuel cells or other microbes. Although discovered ~40 years ago, the EET mechanism remains an unresolved question in environmental microbiology and biogeochemistry. EET via surface-displayed Type 4 pili (T4P), polymers of PilA-N, serving as "nanowires," was hypothesized based on indirect evidence (1) in thousands of papers, from as early as 2002. By correlating genetic, biochemical, and structural studies with EET imaging of live bacteria, we show that PilA-N polymers do not function as nanowires. Instead, our structural, functional, interaction, genetic, and cellular localization studies show that PilA-N binds to another protein, PilA-C, to form PilA-NC filaments that promote secretion of polymeric cytochrome nanowires for EET through an entirely new class of secretion apparatus combining elements of both T4P and Type 2 secretion system (T2SS). These novel findings were facilitated by our suite of complementary methods, enabling the first detailed characterization of the nanowire secretion machinery and identifying proteins involved in secretion and EET. In particular, we describe a novel method to visualize EET in living cells, revealing nanowire composition and mechanism. We combine this approach with genetic and biochemical analyses, an in vivo bacterial two-hybrid assay, atomic force and transmission electron microscopy, and cryo-electron tomography. These detailed biophysical and biochemical studies provide converging evidence that instead of using pili, cells perform EET via cytochrome nanowires that are secreted by a newly identified T4P/T2SS hybrid system. Our studies reconcile decades of contradictory findings and explain the impact of pilA-N mutations on EET by their effect on cytochrome nanowire biogenesis. Our label-free technology for correlated imaging of EET and protein structures can rapidly determine the extent of interspecies electrical connection within complex microbial communities in various environments without the need for cultivation. It could also determine spatial relationships and electrical connections responsible for EET between diverse microbial species and their environment to develop novel, community-based bioenergy strategies. Therefore, it could enable the discovery and engineering of nanowires and their secretion systems in diverse species and environments to determine how various types of nanowires enable one of the most ecophysiologically important microbial processes: anaerobic respiration.

1. Yalcin, S. E., & Malvankar, N. (2020). The blind men and the filament: Understanding structures and functions of microbial nanowires Current Opinion in Chemical Biology, 59, 193-201.

KEYWORDS
extracellular electron trasfer, environmentally important bacteria, cryo-electron microscopy and tomography, atomic and electrostatic force microscpoy, cytochrome secretion system

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HFSP Award Category: Research Grant Program
HFSP Award Year: 2023

REACH OUT AND TOUCH ME: STRUCTURAL AND FUNCTIONAL DIVERSITY OF BACTERIAL AND ARCHAEAL CYTOCHROME OMCS- AND OMCZ-TYPE NANOWIRES REVEALS A WIDESPREAD MECHANISM FOR ANAEROBIC RESPIRATION BY INTERSPECIES ELECTRON EXCHANGE VIA DIRECT PHYSICAL CONTACT

Nikhil Malvankar
Yale University, USA

ABSTRACT
Long-range (>10 µm) microbial extracellular and direct interspecies electron transfer (EET and DIET) drive various global environmental processes and biotechnological applications. Although "nanowires" of polymerized cytochrome OmcS and OmcZ have been proposed to span a long-range EET, their existence beyond Geobacter sulfurreducens (Gs) remains unknown. We show EET and DIET via OmcS-like nanowires by three new species: a thermophilic Geoalkalibacter subterraneus (Ga) isolated from a 1.5-km-deep oilfield; Anaeromyxobacter dehalogenans (Ad) used for bioremediation of toxic materials; and sulfate-reducing bacteria (SRB) Candidatus desulfofervidus auxilii (also called HotSeep-1), which acts as the most efficient (> 80%) biological methane filter by anaerobic oxidation of methane (AOM) via DIET with anaerobic methanotrophic archaea (ANME). ANME also catalyze global biogeochemical cycling of many important nutrients, including nitrogen, phosphate, and iron. Surprisingly, we also found that the first "pure" culture ever obtained for EET-performing ANME 'Candidatus methanoperedens vercellensis' (Mv) produces OmcZ-type nanowires (1). Our findings are novel and significant because methane is 30 times more potent than carbon dioxide in warming the planet, making AOM an important natural process for methane degradation and hence for climate regulation. Despite their global importance, the reaction mechanisms for AOM are poorly understood due to challenges in studying the slow-growing ANME, with doubling times ranging from 1.1 to 7.5 months. A key question is how ANME metabolize at the edge of thermodynamic probability, given the meager energy yield. Remarkably, the AOM rate is 3000 times faster than that accounted for by the diffusion of electron carrier molecules (e.g., H2, formate, acetate). To understand the biochemical and biophysical mechanism of this remarkably fast metabolism, we report the first native structures of these nanowires at unprecedented resolution using cryo-electron microscopy. Mv OmcZ-type nanowire structure reveals key features optimized for rapid? electron conductivity. Combining biochemistry, structural biology, bioinformatics, and imaging, we also identify the assembly machinery and key design principles for OmcS- and OmcZ-type nanowires: (1) reducing heme's solvent exposure decreases their spacing and nanowire aggregation; (2) extensive salt-bridges and PTMs enhance nanowire rigidity and stability to control heme distortion and collinearity; (3) conserved structural features that maintain heme architecture.

This design tunes heme redox potentials and thereby enhances nanowire conductivity to accelerate cell growth. These design principles will also be exploited to develop a new synthetic biology toolbox to rationally engineer on-demand nanowire assembly with tunable electron conductivity in diverse microbial species that lack nanowires to perform EET. Our findings will serve as a blueprint to discover novel species forming nanowires for EET. As structures of OmcS- and OmcZ-type nanowires are conserved and widespread, our studies reveal a specialized biogenesis system for cytochrome nanowires across diverse species and environments.

1. M. Wissink et al., Electrode-associated growth of anaerobic methanotrophic archaea enables structure of OmcZ-type cytochrome nanowire. Preprint https://tinyurl.com/ANME-nanowire, (2026).

KEYWORDS
extracellular electron trasfer, environmentally important bacteria, cryo-electron microscopy and tomography, atomic and electrostatic force microscpoy, cytochrome secretion system

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HFSP Award Year: 2023

MITOCHONDRIAL PEARLING: BIOPHYSICAL MECHANISMS AND BIOLOGICAL FUNCTION

Suliana Manley
EPFL (École Polytechnique Fédérale de Lausanne), Switzerland

Wallace Marshall; Anjana Badrinarayanan; Johan Paulsson
EPFL (École Polytechnique Fédérale de Lausanne)

ABSTRACT
Mitochondrial networks are known to remodel through fission and fusion -- dynamic processes with roles in proliferation, quality control, and complementation. Yet, additional forms of reorganization have been reported, including a “beads-on-a-string” or pearling transition, in which a tubular mitochondrial membrane transforms into regularly spaced, connected beads. Although reported in diverse contexts, the mechanism and function of mitochondrial pearling have remained unclear. In our HFSP team project, we found high-speed light-sheet microscopy that mitochondrial pearling is a widespread and reversible membrane instability. Biophysical theory suggests that it arises from the physical properties of tubular fluid membranes. We observe spontaneous, short-lived pearling events across multiple cell types and physiological states, including T-cell activation, neuronal firing, and replicative senescence. These dynamics reveal two primary classes of physiological pearling, driven by ionic flux or cytoskeletal tension. Through chemical, genetic, and mechanical perturbations, we identify three physical drivers of pearling: osmotic pressure from ionic flux, reduced membrane bending elasticity due to altered membrane packing, and increased membrane tension from external forces. Beyond its biophysical origin, pearling has functional consequences for mitochondrial genome organization. We find that pearling transiently imposes a characteristic length scale along mitochondrial tubules, mediating nucleoid disaggregation and establishing regular spacing between mitochondrial DNA–containing nucleoids with high precision. Calcium influx triggers pearling onset, while the density of lamellar cristae invaginations modulates its prevalence and helps preserve nucleoid spacing after recovery; disruption of these factors leads to aberrant nucleoid clustering. Together, our results unify previously disparate observations of mitochondrial pearling, reveal its physical mechanisms, and identify a role in nucleoid organization. These findings establish pearling as a fundamental aspect of mitochondrial dynamics alongside fission and fusion, with implications for mitochondrial physiology, genome inheritance, aging, and disease.

KEYWORDS
mitochondria, organelle dynamics, membrane instability, nucleoid organization

HFSP Reference Number: RGP0038/2021
HFSP Award Category: Research Grant Program
HFSP Award Year: 2021

BIOLOGICAL CLOCK(S) IN DEEP-SEA MUSSELS

Audrey Mat
University of Vienna, Austria

Federico Scaramuzza[1]; Christophe Klopp[2]; Marjolaine Matabos[3]; Kristin Tessmar-Raible[1]
[1]University of Vienna; [2]University of Toulouse; [3]IFREMER (Institut Français de Recherche pour l'Exploitation de la Mer)

ABSTRACT
The oceans harbour extraordinary biodiversity and are vital to planetary and human health. They regulate our climate via large-scale circulation currents and CO2 sequestration, generate about half of Earth's oxygen via marine phytoplankton, and supply food and energy resources. In fact, 95% of the oceans consists of deep oceans. The deep sea refers to waters below 200m depth, where sunlight is insufficient to support net photosynthesis. The deep sea covers 65% of our planet, yet it is largely unexplored, due to conditions that appear very alien to humans. In the deep sea, hydrothermal vents are habitats that form in tectonic and volcanic areas where fissures induce water circulation in the oceanic crust. The deep-sea mussel Bathymodiolus azoricus is a dominant key ecological species which inhabits hydrothermal vents along the Mid-Atlantic Ridge, at depths ranging -840 to -3350 m, where pressure reaches 84 to 335 bar, and temperatures range from 4°C in ambient seawater to >350°C in black smoker fluids. Internal tides (period 12.4h) modulate the local hydrothermal environment, inducing prominent yet predictable chemical and physical oscillations. Mussels assemblages preferentially occupy cooler areas (4-9°C) and experience alternating influence from the anoxic, acidic, vent fluid enriched in reduced compounds and heavy metals (O_2 = 0 mM, CO_2 = 8 mM, pH 3–3.4, sulphide, Fe, Zn,...), and oxygenated seawater (O_2 ~ 0.2 mM, CO_2 = 2 mM, pH ~7.8). Under ecologically relevant conditions at -1700 m, we previously showed that B. azoricus exhibits biological rhythms - rhythmic valve movements and gene expression - dominated by tidal cycles, with a detectable ~24 h component. Interestingly, under 12:12 light: dark conditions in the laboratory, this dominance reverses. This raised a key question: are these rhythms environmentally driven or governed by one or more endogenous clocks? To address that question, we established molecular and cellular approaches, including primary foot tissue cultures maintained for >3 years, and performed temporal assays that revealed endogenously maintained circadian rhythms under constant conditions. We also identified likely core circadian regulatory elements in B. azoricus, specifically a cross-species conserved non-coding DNA sequence, which includes specific E-box motifs. We validated the transcriptional regulatory activity of this DNA region in heterologous cell assays and showed that it mediates the transcription of the mussel's core circadian clock genes. These results indicate that, at the edge of life, outside solar influence and without known diel signals, animals could use circadian timekeeping to orchestrate their physiology. This has significant implications for our understanding of how deep-sea organisms respond to environmental stimuli, including anthropic disturbances.

KEYWORDS
chronobiology, deep sea, hydrothermal vent, biological rhythms, molecular biology

HFSP Reference Number: RGP021/2024
HFSP Award Category: Research Grant
HFSP Award Year: 2024

WHAT JELLYFISH TEACH US ABOUT TISSUE MECHANICS, SHAPE PROGRAMMING AND ACTIVE MATTER

Anne Materne
Max Planck Institute of Molecular Cell Biology and Genetics, Germany

Zhiqi Shen[1]; Daniel Font-Martín[2]; Vladislav Koren[3]; Ulyana Shimanovich[3]; Chiara Sinigaglia[2]; Carl D. Modes[1]
[1]Max Planck Institute of Molecular Cell Biology and Genetics; [2]Observatoire Océanologique de Banyuls-sur-Mer, CNRS/Sorbonne University; [3]Weizmann Institute of Science

ABSTRACT
Biological shape generation has been studied in a variety of organisms. However, some animals' body plans are extremely complex, making it difficult to disentangle the roles of all individual components involved in tissue shaping processes. In contrast, Cnidarians such as jellyfish are constructed in a seemingly simpler fashion. Yet, they, too, exhibit astounding tissue shaping and regenerative capacities – rendering them great model organisms for tissue level and whole-body investigations. Furthermore, Cnidaria are a sister group to all Bilaterians in the animal tree of life. This means that important evolutionary conclusions can be drawn from processes that either do or do not exist in both groups. Therefore, in our work, we focus on the hydrozoan jellyfish Clytia hemisphaerica. In this organism, the observed rapid wound closure has been implicated to rely, at least partially, on the mechanical properties of the individual tissues in the jellyfish umbrella. In particular, this includes the mesoglea, a thick extracellular matrix structure essential for medusoid shape and body function. Here, based on experimental observations (Sinigaglia lab), we model the mesoglea's and surrounding tissues' roles in wound closure. To this end, we use a coarse-grained spring lattice approach. In this way we can capture essential material processes without knowledge of all molecular- and cellular-scale underpinnings. However, our model also provides the option to incorporate details of cytoskeletal and cellular activity. Hence, we obtain crucial insights regarding how active processes at these scales lead to the emergence of the tissue-level pre-strain in regenerating jellyfish in the first place. Our work shows that simple mesoglea pre-strain is sufficient to initiate closure in a broad spectrum of wound shapes. This finding is in line with the previous experimental work in Clytia hemisphaerica and confirms the essential role of tissue mechanics at the medusa stage of Cnidarian lives. Furthermore, our in-silico model allows us to systematically explore the influence of individual tissues and organs in the animal body's overall mechanical properties. Finally, our results provide unexpected insights for the field of 3D shape-programmable materials. In the future, it will be immensely interesting to explore the role of tissue mechanics in the life history of marine invertebrates more systematically.

KEYWORDS
jellyfish, regeneration, extracellular matrix, spring lattice model, shape-programmable materials

HFSP Reference Number: RGP020/2024
HFSP Award Category: Research Grant Program
HFSP Award Year: 2024

USING MARCHANTIA POLYMORPHA FOR VISUALIZING LIFE IN FOUR DIMENSIONS

Zohar Meir
Gregor Mendel Institute of Molecular Plant Biology, Austria

Magdalena Mosiolek[1]; Thea Kongsted[1]; Soumavo Dolui[1]; Theresa Neubauer[2]; Gabriele Bradamante[2]; Thomas Lendl[2]; Thomas Grentzinger[2]; Astrid Berg[2]; Martin Colombini[2]; Sebastian Seitner[2]; Gerald Schmauss[2]; Alexander Bindeus[2]; Ewa Ksiezopolska[3]; Arnau Sebe-Pedros[3]; Katja Bühler[2]; Liam Dolan[1]
[1]Gregor Mendel Institute of Molecular Plant Biology; [2]Vienna BioCenter (VBC); [3]Barcelona Institute of Science and Technology (BIST)

ABSTRACT
Understanding how individual cells are reprogrammed within a whole organism is essential to deciphering the dynamic processes driving diverse morphogenetic outcomes. Historically, textbook models of morphogenesis have focused on the highly reproducible embryonic development in species like Drosophila or C. elegans. However, for many less canonical processes—such as tumor progression or tissue regeneration—the instantaneous states at early stages do not reliably predict the final outcomes. Capturing these trajectories requires continuous 3D recordings to eliminate biased inferences from static snapshots. However, obtaining such data remains challenging in practice, as 4D imaging is often restricted to surface layers, and its scalability is hampered by manual sample maintenance and image processing. To overcome these challenges, we developed an automated 4D imaging platform to track the development of Marchantia polymorpha plants from single-celled spores. Naturally released from the mature plant and developing in isolation, spores serve as a unique model for scalable, quantitative analysis of uninterrupted whole-organism morphogenesis. Using a machine-learning pipeline, we mapped cell ontogeny in hundreds of individual plants and classified cells by their morphogenetic signatures, nuclear shape, and chlorophyll composition. This allowed us to characterize the stepwise formation of the first differentiated cell—the highly elongated rhizoid cell—and demonstrate how impaired rhizoid cell development affects neighboring cells, illustrating how constant cell-cell communication shapes the Marchantia plant from its earliest developmental stages. To resolve the underlying molecular mechanisms that operate during the development of the spore into the mature plant, we generated over 500,000 single-nucleus RNA-seq profiles across 24 time points, charting the transient transcriptional states specifying cell types and their differentiation trajectories. Our integrated analysis revealed a transient state in spores where light cues activate cell-cycle programs, providing a novel mechanistic link between environmental signals and genomic growth control. We further characterized a 30-minute "stem-like" state following cell division, providing a rare view of the transcriptional dynamics taking place during the formation of the inner cell plate—a short-lived syncytial-like phase and a hallmark of plant cytokinesis. Finally, we show that while all spores eventually establish a stem-cell niche, the timing and morphogenetic paths are highly variable.

By analyzing the transcriptomes and imaging of over 1,000 individual plants, we characterize this variance and uncover a molecular switch acting before meristem initiation. Discovering how the first stem-cell niche forms in plants will be a key to understanding their later development and to testing how stochastic biological systems converge into the robust formation of stable niches with particular, multipotent characteristics.

KEYWORDS
Image Analysis; Developmental Genetics; Bioinformatics; Single-cell Genomics; Plant biology

HFSP Reference Number: LTF0024/2023
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2023

ENHANCING TROPICAL TREE-RING DETECTION USING A SEMI-AUTOMATED SWIR HYPERSPECTRAL IMAGING PIPELINE

Meley Mekonen Rannestad
Norwegian University of Life Science (NMBU), Norway

Muhammad Hassaan Farooq Butt[1]; Olusegun Olaitan Akinyemi[1]; Abebe Damtew Awraris[2]; Buruh Abebe Tetemke[2]; Zenebe Girmay Siyum[2]
[1]Norwegian University of Life Sciences (NMBU); [2]Mekelle University

ABSTRACT
Dendrochronological methods are well established in temperate and boreal regions, where strong seasonal cycles produce clearly defined annual rings that can be directly related to climatic variability. In tropical environments, however, the weaker or irregular expression of seasonality often results in discontinuous or irregular growth boundaries, making ring identification challenging and limiting the reliability of climate reconstructions. While many tropical species respond to rainfall or temperature seasonality, the resulting ring boundaries frequently lack sufficient contrast in the visible spectrum (RGB; 400–700 nm), reducing the effectiveness of conventional scanning and manual annotation approaches that rely on optical contrasts between earlywood and latewood. To address this limitation, we present a semi-automated pipeline that leverages Short-Wave Infrared (SWIR; 1000–2500 nm) hyperspectral imaging to enhance the detection of ring boundaries in tropical hardwoods. Instead of relying solely on anatomical features visible to the eye, our approach exploits chemical-specific reflectance signatures inherent to wood composition. Hyperspectral data cubes were acquired from discs of Juniperus procera, Olea europaea subsp. cuspidata, Faidherbia albida, and Vachellia lahai. Principal Component Analysis (PCA) applied to targeted regions of interest effectively separated anatomical structure from surface noise, with the first component reliably accentuating ring boundaries that appeared ambiguous in RGB imagery. To facilitate quantitative analysis, circular discs were converted to a polar coordinate system using an automated geometric transformation, followed by a sector-based signal-processing framework. Gaussian detrending was incorporated to mitigate large-scale trends associated with heartwood–sapwood transitions, enabling a more consistent and reproducible preliminary ring count. Across all tested specimens, SWIR-derived contrast maps revealed structural patterns that remained undetectable through manual inspection alone. This pipeline provides a powerful complementary tool for tropical dendrochronology, offering improved boundary visibility, reduced subjectivity in annotations, and a more reproducible basis for subsequent high-resolution measurements in species where traditional ring identification has historically been problematic.

KEYWORDS
Short-wave infrared hyperspectral imaging (SWIR), Tropical dendrochronology, Tree-ring detection, Polar coordinate transformation, Principal Component Analysis (PCA)

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HFSP Award Category: Research Grant Early Career
HFSP Award Year: 2024

THE ROLE OF LIPID PHYSICAL PROPERTIES FOR THE MULTIFUNCTIONALITY OF INSECT CUTICULAR HYDROCARBONS

Florian Menzel
ohannes Gutenberg University Mainz, Germany

Walter Federle[1]; Fumitoshi Kaneko[2]
[1]University of Cambridge; [2] Kyushu University

ABSTRACT
Insects wear a multifunctional suit on their body surface - a thin lipid film consisting of cuticular hydrocarbons (CHCs). This outermost layer of the insect's body has a complex chemical composition that varies drastically across species, consisting of unbranched and branched alkanes as well as alkenes and/or alkadienes. It not only serves as vital barrier against desiccation, but also acts as one of the most important communication signals for insects. CHCs also have important biomechanical functions such as surface adhesion and joint lubrication. All these biological functions crucially depend on the physical properties of the CHC films, which are still poorly understood. How can this layer fulfil so many functions simultaneously? How did the enormous diversity of CHC compositions across insects evolve? These central questions are still largely unexplored. Out team studies the phase behavior, melting ranges, molecular arrangement, diffusion rates and viscosity of cuticular hydrocarbons, and how these depend on their chemical composition, by using diverse methods ranging from synchrotron X-ray scattering, Fourier transform infrared (FTIR) spectroscopy with transmission, reflection, ATR, infrared microscopy, and polarization analysis as sampling techniques to particle tracking micro-rheology, thermal desorption and gas chromatography-mass spectrometry. We want to understand how chemical composition determines physical properties and in this way explain their biological functions. In-situ synchrotron X-ray scattering measurements on several insect species revealed orthorhombic polyethylene-type packing of saturated hydrocarbons on all cuticular surfaces. Observation of temperature-dependent X-ray scattering revealed that CHCs occur as a mixture of solid and liquid phases, and the solid proportion increases with decreasing temperature. The X-ray reflections due to the lateral hydrocarbon packing indicate that branched and unsaturated CHCs have a looser lateral packing than saturated, straight-chain CHCs. This property enables investigating the behavior of unsaturated and branched hydrocarbons separately from straight-chain alkanes. Phase separation between different hydrocarbons was confirmed not only by differential scanning calorimetry (DSC) measurements on insect cuticular extracts and artificial hydrocarbon mixtures, but also by in-situ DSC measurements on insect wings. Micro-rheology of cuticular hydrocarbon extracts showed that the liquid phase has a bimodal viscosity distribution, with low-viscosity (similar to olive oil) and high-viscosity (similar to honey) domains. This separation may be crucial in avoiding conflicts between waterproofing (which requires high-viscosity compounds) and communication (which requires low-viscosity compounds).

Force measurements on insect cuticle showed that friction is reduced in the presence of cuticular hydrocarbons, giving rise to very low friction coefficients. Experimental removal of cuticular hydrocarbons resulted in higher friction forces, and an even stronger increase in shear stress (friction force per contact area), highlighting the importance of hydrocarbons for lubricating the insects' body surface and joints. Taken together, our results demonstrate that the physical properties of cuticular hydrocarbons are crucial for their various biological functions.

KEYWORDS
cuticular hydrocarbons, phase behaviour, ecophysiology, insects, multifunctionality

HFSP Reference Number: RGP008/2023
HFSP Award Category: Research Grant
HFSP Award Year: 2023

DECODING THE MECHANISMS OF CONDENSATE-MEMBRANE INTERACTIONS IN CHANGING CELLULAR ENVIRONMENTS

Dragomir Milovanovic
Charite -Universitätismedizin Berlin, Germany

Johannes Vincent Tromm[1]; Kazuki Tsujimura[2]; Ana Kodra[3]; Simon Leitner[4]; Meline Macher[5]; Vivian Lobo[6]; Meng-Meng Fu[7]; Kathrin Leppek[6]; Aleksandar Matkovic[4]; Anthony Hymann[5]; Simon Alberti[8]; Yusuke Hirabayashi[2];
[1]Charite -Universitätismedizin Berlin; [2]University of Tokyo; [3]German Center for Neurodegenerative Diseases (DZNE); [4]Technical University of Leoben; [5]MPI-CBG; [6]UK Bonn; [7]UC Berkeley; [8]TU Dresden

ABSTRACT
Recent discoveries have revealed the dynamic interplay between membrane-less organelles—known as biomolecular condensates—and membranes, highlighting their crucial role in coordinating cellular activities such as neurotransmitter release. We investigate the condensate-membrane interaction in cellular physiology using G3BP1/2 (G3BP) proteins, which are essential for integrating metabolic response to changing nutrients and cellular stress. Our data indicate that lipid composition determines the level of interaction between G3BP condensates and membranes. At the lysosomal surface, G3BP act as tethers for the to restrain mTORC1 signaling and couple metabolic status to stress-adaptive growth control. In the cytoplasm, G3BP function as nucleators of stress granules, promoting condensation of non-translating mRNPs into RNA-rich granules that protect transcripts and remodel gene expression programs during the integrated stress response. However, how cells execute the switch from a lysosomal nutrient sensor to a cytosolic mRNA restrainer function of G3BP remains unknown. Our data indicate the structural role of RNA molecules in modulating the viscoelastic properties of G3BP condenses leading to membrane de-wetting and favoring RNA granule formation. Using a newly established 2D graphene sensors, we discovered that G3BP condensates accumulate electric potential at their interfaces, suggesting the biomolecular condensates as the putative mesoscale capacitors. In fact, the presence of RNA leads to reduction of interfacial electric double layer, indicating the dependance of condensates from local ion concentration. Together, our data indicate how material properties of bimolecular condensates as electrical sensors allow for the changing environmental conditions to switch the signaling cascades. Through these dual activities as lysosomal signaling scaffolds and condensate-forming RNA chaperones, G3BP condensates couple nutrient sensing to selective mRNA silencing to enforce an adaptive stress program.

KEYWORDS
phase separation, RNA, metabolism, stress granules, graphene sensors

HFSP Reference Number: RGEC32/2023
HFSP Award Category: Research Grant Early Career
HFSP Award Year: 2023

CHARTING THE ORIGIN, DIVERSITY AND BIOGEOGRAPHY OF SMALL PROTEINS IN THE GLOBAL OCEAN MICROBIOME

Samuel Miravet-Verde
ETH Zurich, Switzerland

ABSTRACT
Emerging evidence shows that small open reading frame (smORF)-encoded proteins (SEPs; ≤100 aa) are key regulators of microbial physiology and competition, yet they remain largely overlooked in genome annotations, representing a vast hidden layer of functional diversity. Their small size enables rapid regulatory responses and frequent roles in antimicrobial activity and molecular interactions, making them central to microbial ecology and evolution. The ocean, with its immense microbial diversity, strong environmental gradients, and intense resource-driven competition, is a prime reservoir for SEP diversification. However, fragmented genomic datasets and size-biased annotation pipelines have prevented a comprehensive understanding of SEP diversity, distribution, and ecological roles in marine systems. To address this, we established the Ocean Microbiomics Database (OMDB; https://omdb.microbiomics.io/) by uniformly reconstructing and curating >274,000 metagenome-assembled genomes (MAGs) from >12,000 marine samples, representing >30,000 prokaryotic species. We paired this resource with mSEPs, a deep-learning framework leveraging physicochemical and structural properties of SEPs to discriminate coding from non-coding smORFs, enabling high-throughput prediction across MAGs. Using this approach, mSEPs identified 298 million unique smORF sequences with high coding potential, representing an unprecedented expansion of the predicted marine SEP repertoire. Functional predictions further revealed 81,892 sequences with potential antimicrobial activity (AMPs), highlighting both their ecological and biotechnological significance. Notably, the cyanobacterium Trichodesmium erythraeum emerged as a prominent producer, with 16 predicted AMPs, 12 of which are potentially hemolytic, providing a possible molecular explanation for fauna mortality during its blooms. Leveraging the enriched metadata from OMDB, we uncovered ecological patterns linking SEP frequency to ocean temperature via GC content dynamics. This trend also extends to AMPs, suggesting that ocean warming may promote increased de novo generation of small proteins and AMPs, with the potential to reshape community dynamics. Comparative genomics further indicates that smORF fusion can drive the evolution of larger proteins, with some clades showing up to 15% of proteins derived from fusion events, underscoring their role in protein innovation. Together, these findings establish the global ocean as a vast functional reservoir of uncharacterized SEPs. By studying them at a large-scale, genome-resolved level, this work reveals their central roles in microbial systems and demonstrates how small proteins shape key eco-evolutionary processes.

KEYWORDS
ocean, microbiome, small proteins, antimicrobials, bioinformatics

HFSP Reference Number: LT0050/2023-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2023

THE ORIGIN OF SYNAPTIC NEUROTRANSMISSION IN ANIMALS

Jacob Musser
Yale University, USA

Aissam Ikmi[2]; Rong Xuan Zang[1]; Shigeki Watanabe[3]
[1]Yale University; [2]European Molecular Biology Laboratory (EMBL) Heidelberg; [3]Johns Hopkins School of Medicine

ABSTRACT
Synaptic neurotransmission evolved early in animals, enabling rapid, spatially precise intercellular communication to coordinate behavior and physiology across the body. To understand how this emerged, we study sponges and sea anemones, two early-diverging lineages that bracket the origin of synaptic transmission. Sponges are filter-feeding animals that lack neurons and synapses yet coordinate stereotyped contraction and relaxation of their water canal system in response to agitation and environmental cues. Using a broad pharmacological screen, we find that sponges deploy a diffusion-based signaling system centered on monoamines, including tryptamine, phenethylamine, and tyramine. These transmitters are synthesized and packaged by noncanonical biosynthetic enzymes and vesicular transporters, and are released locally by distributed sensory-secretory cells within different regions of the water canal system. Using phosphoproteomics, we show tryptamine acts through GPCRs coupled to RhoA/ROCK signaling to drive actomyosin-dependent remodeling in canal contractile epithelia. In sea anemones, we are using fine-tuned protein language models to infer protein–protein interactions and reconstruct the evolutionary assembly of presynaptic scaffolds. As an initial example, our preliminary predictions identify an ancient active-zone scaffold involving RIM proteins that may couple calcium channel localization to vesicle release machinery. Together, these findings suggest early animal nervous systems evolved from localized, diffusion-based signaling to synaptic transmission via new junctional scaffolding interactions that enabled fast, long-distance, synchronized communication across animal bodies.

KEYWORDS
nervous system, evolution, synapse, neurotransmission, animal multicellularity

HFSP Reference Number: RGP024/2023
HFSP Award Category: Research Grant Program
HFSP Award Year: 2023

DISSECTING DIRECTIONAL AND LATERAL DYNAMICS IN INTRACELLULAR CARGO TRANSPORT USING MINFLUX

Farha Naaz
University of Warwick, UK

Alex Fellows[2]; Richard Wademan[2]; Anne Straube[1]
[1]University of Warwick; [2]MRC-LMB

ABSTRACT
Cellular cargoes, such as organelles and secretory vesicles, recruit molecular motors (kinesin and dynein) for transport along microtubules. Quantitative dissection of these behaviours is essential for understanding how motors coordinate long-range transport within crowded cellular environments. Bidirectional transport is crucial for neuronal function, and defects or mutations in motor-dependent transport have been linked to neurodegenerative diseases. These motors exhibit dynamic behaviours including processive runs, pauses, directional reversals, and potential switching between microtubule tracks or protofilaments. Quantitative resolution of these transitions is critical for understanding how motor coordination regulates cargo distribution. Using MINFLUX, a super high-resolution imaging technique, we investigate dynein and kinesin dynamics in neurons to understand their engagement with microtubules at high spatiotemporal resolution. We utilize inducible neurons expressing endogenously Halo-tagged motors (dynein, kif5a, and kif5c) with low-level labelling (2–100 pM) of Janelia Fluor Halo dyes to track individual fluorophores. By analysing XY trajectories of processively moving motors over time, we classify runs as diffusive, static, or actively moving based on velocity. To assess directional speed differences, we use EB3-mStayGold (EB3-mSG) as a plus-end microtubule marker, distinguishing actively moving motors from hitchhikers. Molecular motors frequently exhibit sideways stepping and reversals, making time resolution crucial for accurate trajectory analysis. To better capture true reversals and jumps, we applied smoothing techniques to the data. On-axis displacements enable robust quantification of run lengths, dwell times, and reversal frequency. Simultaneously, off-axis displacement dynamics were analysed to detect abrupt lateral excursions consistent with microtubule switching or protofilament transitions. The precision we achieved ranged from 0.5–2.5 ms in time and 4–8 nm in the X and Y directions. By resolving fine-scale dynamic transitions within continuous trajectories, this approach advances our understanding of how molecular motors orchestrate regulated intracellular transport.

KEYWORDS
Kinesin, Dynein, Microtubules, MINFLUX microscopy, Single-molecule tracking

HFSP Reference Number: LT0044/2023
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2023

DECODING ENHANCER-MEDIATED GENE REGULATION DURING MALE GERMLINE DEVELOPMENT

Masahiro Nagano
Massachusetts Institute of Technology (MIT), USA

ABSTRACT
Enhancers are primary regulators of cell-type-specific gene expression, and their misregulation contributes to developmental disorders and cancer. Yet quantifying enhancer function remains challenging: regulatory effects can span long genomic distances, is highly cell-type-specific, and often reflects combinatorial control, with multiple enhancers converging on a single gene. Consequently, we still lack a quantitative framework to estimate the effect size of individual enhancer regions on transcription. A major technical barrier is that conventional 3C assays (e.g., Hi-C) often miss or blur enhancer–promoter interactions (EPIs), limiting accurate measurement of enhancer–promoter contact frequencies. To overcome this limitation, we leveraged Region-Capture Micro-C (RCMC), an ultra–high-resolution 3D genomics method developed in the Hansen lab that achieves ~100-fold higher effective resolution than prior 3C approaches. Using a scalable in vitro mouse male germline differentiation system, we generated ultra-deep Micro-C and RCMC maps across key stages, revealing ultra–fine-scale 3D genome reorganization and enabling quantitative measurement of stage-specific enhancer–promoter contact frequencies. In parallel, we profiled scMultiome (joint scRNA-seq and scATAC-seq) across 11 time points to nominate candidate cis-regulatory elements genome-wide. We are integrating these datasets with the scE2G framework to infer cell-type-specific enhancer–gene links and to assemble a putative enhancer atlas across germline development. Finally, we established single-enhancer knockout lines targeting ~30 candidate enhancers across multiple loci, quantified ~1,040 cis-regulatory interactions, and developed a preliminary quantitative model to predict transcriptional effect sizes. Together, our ultra–high-resolution 3D genomics resolves 3D genome reorganization during germline differentiation at unprecedented resolution and provides a quantitative foundation for understanding enhancer–promoter regulation.

KEYWORDS
Germ cell development, 3D genome organization, Enhancer-promoter regulation

HFSP Reference Number: LT0022/2024-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2024

MEAT, MINDS, AND MOLECULES: OVERCOMING FUNDAMENTAL ANALYTICAL CHALLENGES IN ISOTOPIC MEASUREMENTS OF FOSSIL AMINO ACIDS

Caj Neubauer
University of Colorado Boulder, USA

Leonhard Prechtl[2]; Rani Bakkour[2]; Florian Rubach[3]; Alfredo Martínez-García[3]; Jana Storsberg[3]; Jennifer Leichliter[3]; Matt Sponheimer[1]; Daryl Codron[4]; Dwight E. Matthews[5]; Tina Lüdecke[3]
[1]University of Colorado, Boulder; [2]Technical University of Munich; [3]Max-Planck Institut für Chemie; [4]University of the Free State; [5]University of Vermont

ABSTRACT
Dietary change, particularly the incorporation of nutrient-rich animal resources, is widely believed to have fueled the evolutionary expansion of the human brain. However, direct evidence of when early hominins made this transition remains scarce. Traditional stable isotope analysis relies on the maxim "you are what you eat," using bulk measurements in fossils to infer trophic levels. Yet, this approach often lacks the specificity to distinguish complex nutrient routing or specific food sources. To address this limitation, we pursue a methodological advance: moving from bulk measurements to compound-specific isotope analysis (CSIA) of individual amino acids preserved in fossil tooth enamel. However, because analysis of trace amounts of amino acids in ancient geological records remains in its infancy, our HFSP-supported collaboration is developing a novel analytical approach to make this unique molecular archive accessible. Through sensitive biogeochemical assays we find increasing evidence that amino acids are preserved in fossil tooth enamel on the timescales of millions of years. To isolate these trace quantities from the complex mineral apatite matrix, we are developing highly selective extraction protocols using molecularly imprinted polymers (MIPs). While our targeted approach successfully captures the analytes, we are currently optimizing the protocol to further decrease column bleed, which we found causes critical interference with downstream mass spectrometry. For the isotopic measurements, we focus on the differences between glutamic acid, a non-essential amino acid acting as a nitrogen reservoir, and phenylalanine, an essential amino acid carrying the nitrogen isotopic signature of the diet. We utilize electrospray-ionization Orbitrap isotope ratio mass spectrometry (ESI-Orbitrap IRMS) to achieve the necessary molecular isotopic resolution and sensitivity. With this approach, we can measure 13C down to the low-picomolar sensitivity range. However, 15N measurements – which are highly desirable for reconstructing meat consumption – exhibited unexplained noise. To overcome this limitation, we have started to better understand the fundamental physical chemistry of ESI and its associated isotope effects. Achieving high-precision isotope ratios at this extreme sensitivity also requires very robust isotopic referencing protocols; thus, we are exploring innovative approaches to fundamentally enhance the precision and accuracy of ESI-Orbitrap IRMS.

By confronting these fundamental analytical challenges, this project helps move hominin paleodietary reconstruction beyond basic trophic level assignments toward revealing more nuanced foraging behaviors. Furthermore, because isotope ratio analysis is a unique tool for many areas of science, the new technology we develop directly catalyzes progress in diverse fields. As examples, we will briefly highlight recent contributions to climate research and the study of environmental pollutants.

KEYWORDS
Paleo Diet, Stable Isotopes, Amino Acids, Molecularly Imprinted Polymers, Mass Spectrometry

HFSP Reference Number: RGP019/2023
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HFSP Award Year: 2023

UNRAVELLING THE MECHANISM OF SCHISTOSOME EGG MIGRATION IN A COMPLEX HOST ENVIRONMENT

Lucy Ochola
Centre for Research and Therapeutics and Institute of Primate Research, Kenya

Kennedy Okeyo[1]; Martin Omondi[2]; Brianna Corman[1]; Paul Ogongo[3]; Dr Euiwon Bae[1]; Paul Robinson[1]

[1]Purdue University; [2]University of Nairobi and Institute of Primate Research; [3]University of California, San Francisco

ABSTRACT
Schistosoma mansoni, the causative agent of intestinal schistosomiasis, remains a major public health challenge in tropical regions. A central enigma of infection is the migration of parasite eggs despite lacking intrinsic motility through host tissues to the intestinal lumen for excretion. While endothelial cells have been implicated in this process, the contribution of other cell types, particularly fibroblasts, is poorly understood. Moreover, the factors determining whether eggs successfully reach the gut lumen or become trapped in tissues, leading to granuloma formation and pathology, remain unclear. Elucidating these mechanisms may reveal new strategies for controlling S. mansoni infection and associated disease burden. This study aims to investigate egg migration within the complex host environment by integrating analyses of tissue remodeling, immune responses, and egg gene expression during infection. To clarify host immune responses at different phases of infection, a longitudinal study was performed. Mice were infected with S. mansoni cercariae and sampled weekly for fourteen weeks to assess the kinetics of key cytokines associated with dynamic host-parasite interactions across infection stages. Th1/Th2/Th17 associated cytokines (IL-4, IL-10, TNF-α, IFN-γ, IL-33, IL-17) were profiled using Luminex and further correlated with immune cell populations (CD4, CD8, CD25, CD 31, CD 68) that were assessed via flow cytometry. We found that cytokine levels tend to fluctuate as infection progresses, with key Th2 cytokines (IL-4 and IL-10) increasing during the chronic phase of disease. These cytokine levels could be associated with T cell differentiation and immune regulatory responses. To visualize cellular interactions and tissue remodeling associated with granuloma formation, studies are ongoing to examine how the remodeling of the extracellular matrix occurs in the liver tissues around migrating eggs using immunohistochemistry, and to establish how the remodeling correlates with the expression of fibroblast-related genes (FSP1, FAP, Fibronectin, α-SMA) to be quantified using quantitative PCR. Together, these approaches will unravel the cellular and molecular mechanisms that govern egg migration and entrapment, advancing our understanding of schistosomiasis pathology and potential avenues for intervention.

KEYWORDS
Schistosomiasis, fibroblasts, cytokines, granuloma, gene expression

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HFSP Award Category: Research Grant
HFSP Award Year: 2023

IMMUNITY AND SELFISHNESS IN GENOMIC VARIABILITY HOTSPOTS OF ARABIDOPSIS THALIANA

Gal Ofir
Max Planck Institute for Biology Tübingen, Germany

Anna Cavalieri; Lukas Burkhardt; Sicheng Xu; Naja Faerber; Sandra Mingo-Ramirez; Kevin D. Murray; Detlef Weigel
Max Planck Institute for Biology Tübingen

ABSTRACT
Host-pathogen interactions are powerful drivers of rapid evolution, shaping some of the most diverse and complex loci in species pangenomes - a pattern conserved across bacteria, animals, and plants. In the model plant Arabidopsis thaliana, highly variable genomic regions are known to harbor NLR (nucleotide-binding leucine-rich repeat) immune receptor genes. What additional gene families and genomic phenomena are hidden in these rapidly evolving regions? To address this, we systematically analyzed the genomic neighborhoods of NLR genes across a curated collection of high-quality A. thaliana genomes from the 1001G+ Project. The complex and repetitive NLR clusters, together with their flanking regions, are fully resolved in these assemblies. We identified several non-NLR gene families with high pangenomic variability not previously reported that are consistently localized in NLR genomic neighborhoods. Their genomic signature and predicted functions position them as strong candidates for novel components of plant immunity. Leveraging the same high-quality assemblies, we investigated additional high-complexity genomic regions and uncovered extreme duplication and translocation events of essential genes into centromeric regions. We show that these accession-specific centromeric duplication clusters are transcriptionally active, and are associated with epigenetic silencing of the canonical reference copies - rendering individual plants dependent on these centromeric duplicates for expression of essential genes. Together, these findings illustrate how fully resolved genome assemblies can serve as a powerful tool for discovering unexpected dimensions of plant biology.

KEYWORDS
plant science,genomics,immunity,selfish elements

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HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2022

ILLUMINATING ORPHAN GPCR SIGNALING BY NOVEL BIOCHEMICAL APPROACHES

Hiroyuki Okamoto
University of California, San Francisco, USA

Aashish Manglik
University of California, San Francisco

ABSTRACT
G protein-coupled receptors (GPCRs) are important for cells to sense their external environment. Studies on their structure, pharmacology, and so on have led to successful therapies of GPCR-related diseases. However, there are still hundreds of «orphan» GPCRs, whose function remain unknown. Revealing the function and ligands of orphan GPCRs is called “deorphanization”. Deorphanization of these receptors could have significant implications for human physiology and disease treatment, but efforts to find their activating molecules have slowed down over the past decade. The lack of an activator makes it difficult to study downstream signaling pathways and to develop assays for probe molecule discovery of orphan GPCRs. My research aims to develop new approaches to illuminate the function of orphan GPCRs, particularly those expressed in the central nervous system. To overcome these challenges, I propose a new approach to purify orphan GPCRs and use them as ”bait” to find endogenous hormones or surrogate activators for orphan GPCRs by combining such disciplines as biochemistry, native mass spectrometry, antibody development, and structural biology. As a prototype of screening of orphan GPCR activators, I chose GPR85, the most conserved GPCR in all vertebrates. Knock out of GPR85 leads to a 20% increase in brain mass, while overexpression studies implicate it as a negative regulator of hippocampal neurogenesis and spatial learning. But the lack of ligand of GPR85 hinders understanding of the precise function and signaling of GPR85. My research has two aims. First, identify endogenous ligands for GPR85 using mass spectrometry and NMR techniques. To accomplish this, I will use extracts from the hippocampus to identify other molecules that bind tightly to GPR85. The identified molecules will be used to determine the cellular signaling pathways modulated by GPR85. Second, use combinatorial antibody libraries to find new surrogate ligands for GPR85. The approach will identify molecules that bind to GPR85 in a specific conformation and have the potential to induce or inhibit GPR85-mediated signaling. The candidate molecules will be screened for activity against a panel of cellular signaling pathways. Orphan GPCRs represent a huge opportunity for drug discovery. A new approach could shed light on the function of these mysterious receptors and pave the way for similar approaches to tackle other orphan receptors. The ultimate goal is to illuminate the function of one of the most intriguing orphan GPCRs in the genome and pave the way for similar approaches to numerous other orphan receptors. This approach could lead to the discovery of new therapies for recalcitrant diseases.

KEYWORDS
GPCR, Deorphanization, GPR85, IP-MS

HFSP Reference Number: LT0011/2024-L
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HFSP Award Year: 2024

UNRAVELLING THE MECHANISM OF SCHISTOSOME EGG MIGRATION IN A COMPLEX HOST ENVIRONMENT

Kennedy Okeyo
Purdue University, USA

Lucy Ochola[2]; Martin Omondi[3]; Brianna Corman[1]; Paul Ogongo[5]; Euiwon Bae[1]; J. Paul Robinson[1]
[1]Purdue University; [2]Kenya Institute of Primate Research; [3]University of Nairobi; [5]University of California, San Francisco

ABSTRACT
Schistosoma mansoni, the causative agent of intestinal schistosomiasis, remains a major public health challenge in tropical regions. A central enigma of infection is the migration of parasite eggs through host tissues to the intestinal lumen for excretion; a puzzling process that occurs even though Schistosoma eggs lack intrinsic motility mechanisms. While endothelial cells have been implicated in the migration process, the contribution of other cell types is poorly understood, including fibroblasts that are well known for their contractile property and immune stimulation. By integrating egg-host interaction and tissue mechanics, this study aims to quantify and dissect the nature and origin of forces that propel schistosome egg migration through the complex host tissues, using both in vitro and animal models. Since investigating complex biological processes in vivo is challenging, we are establishing in vitro systems using stiffness-tuned hydrogels to mimic the in vivo microenvironment toward recapitulating the migration events in vitro. To this end, we have designed and developed a cost-effective, in-house gel stiffness characterization platform capable of simultaneously measuring stress and strain, enabling us to estimate and fine-tune the material properties of hydrogels such as stiffness (Young's modulus). Moreover, to map forces associated with egg migration based on a tissue mechanics approach, a computational framework has been developed in MATLAB that enables mapping of microscale deformations associated with Schistosoma egg migration in an in vitro system. The algorithm detects pixel-level displacements in sequential microscope images using two independent image-correlation techniques: cross-correlation and phase-shift correlation, yielding a displacement field from which forces can be quantified when the matrix stiffness is predetermined. Furthermore, to gain in vivo insights, our co-investigator has established infected mouse models to assess longitudinally the kinetics of key cytokines as a readout of immune response associated with egg-host interaction. By further correlating the data with T-cell differentiation and kinetics, we are discerning host immune responses across different infection phases, and associating them with granuloma formation, and tissue remodeling which accompany egg deposition and excretion. Thus, by integrating in vitro and in vivo studies, we are dissecting what propels schistosome egg migration. Elucidating this mechanism may inform new strategies for controlling Schistosoma infection and associated disease burden, with immense health and economic benefits.

KEYWORDS
Schistosoma egg migration, granuloma, immune response, tissue mechanics, in vitro systems

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HFSP Award Category: Research Grant
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MATLAB-BASED PARTICLE TRACKING ALGORITHM FOR SCHISTOSOMA EGG MIGRATION ANALYSIS

Kennedy Okeyo
Purdue University, USA

Carlos Felix[1]; David Kim[1]; Brianna Corman[1]; J. Paul Robinson[1]; Paul Ogongo[2]; Martin Omondi[3]; Lucy Ochola[4]; Euiwon Bae[1]
[1]Purdue University; [2]University of California, San Francisco; [3]University of Nairobi; [4]Kenya Institute of Primate Research

ABSTRACT
Schistosomiasis is a globally prevalent parasitic infection characterized by the migration of Schistosoma eggs through host tissues—a mechanically driven process that remains insufficiently understood. Conventional investigative approaches, including epidemiological mapping, immunological assays, and organ-on-a-chip platforms, provide important biological insights but lack the capacity to directly quantify microscale tissue deformations induced by host-mediated forces. To address this limitation, a computational framework was developed in MATLAB for potential mapping of microscale deformations associated with Schistosoma egg migration in an in vitro system being developed to mimic in vivo migration events. The framework's algorithm detects pixel-level displacements in microscopy images using two independent image-correlation techniques: cross-correlation and phase-shift correlation. Algorithm performance was validated through controlled simulations in which predefined and random pixel displacements were applied to 256 × 256 grayscale images partitioned into grids of varying resolutions. Both correlation methods achieved 100% accuracy in 4 × 4 grid analyses and demonstrated high accuracy (90–100%) in 8 × 8 grid configurations, confirming their reliability under idealized test conditions. A complementary vector-plotting routine was implemented to visualize displacement fields and translate computational outputs into interpretable representations. Collectively, these findings establish image-based correlation analysis as a robust and sensitive approach for quantifying subtle deformation, providing a technique for quantifying microscale forces associated with egg migration. Future work will incorporate real-time microscopy images of Schistosoma eggs embedded within hydrogel scaffolds—mechanically tunable substrates that recapitulate tissue-like environment—to quantify the contribution of host cells to egg translocation. Beyond schistosomiasis research, this computational strategy holds broader applicability in soft-tissue biomechanics and mechanobiology.

KEYWORDS
Schistosoma egg migration, tissue deformation, in vitro system, fluorescent microscopy, image-correlation analysis

HFSP Reference Number: RGP020/2023
HFSP Award Category: Research Grant
HFSP Award Year: 2023

DESIGN, FABRICATION, AND TESTING OF INTEGRATED GEL STIFFNESS CHARACTERIZATION SYSTEM FOR EVALUATING SCHISTOSOME EGG MIGRATION

Kennedy Okeyo
Purdue University, USA

David Kim[1]; Aditya Brahmbhatt[1]; Brianna Corman[1]; J. Paul Robinson[1]; Paul Ogongo[2]; Martin Omondi[3]; Lucy Ochola[4]; Euiwon Bae[1]
[1]Purdue University; [2]University of California, San Francisco; [3]University of Nairobi; [4]Kenya Institute of Primate Research

ABSTRACT
Investigating the interactions between Schistosoma eggs and host tissues, particularly the forces propelling their migration through the complex host microenvironment is extremely challenging in vivo. To this end, in vitro models established with extracellular matrix (ECM) can be useful to recapitulate egg translocation and permit direct visualization and quantitative analysis of the complex migration processes. The present project aims to design and develop a cost-effective, in-house gel characterization platform capable of simultaneously measuring stress and strain, thereby enabling estimation of material properties such as Young's modulus. The system architecture incorporates a 3D-printed polylactic acid (PLA) frame and laser-cut black acrylic housing, along with interchangeable 2D grid-patterned fixtures for secure sample mounting. Mechanical extension is generated via a stepping motor, which applies controlled uniaxial deformation to an ECM mimicking hydrogel specimen to induce measurable stress and strain. A load cell records normal force, while an upright fluorescence microscope tracks embedded fluorescent particles (beads) to quantify strain through displacement analysis. System control and data acquisition are managed by a single Arduino microcontroller interfaced with a laptop computer. Compared to a commercial rheometer, this integrated platform enables low-cost, quantitative assessment of gel stiffness and related mechanical properties. Ultimately, it supports the broader objective of elucidating the role of material mechanics in facilitating Schistosoma egg propulsion within a physiologically relevant in vitro system that are being developed to mimic in vivo egg migration events.

KEYWORDS
Schistosoma egg migration, in vitro system, hydrogel stiffness characterization, fluorescent microscopy, 3D printing

HFSP Reference Number: RGP020/2023
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TRANSIENT DORSAL PHARYNGEAL CAVITY IN MEDAKA LARVAE REVEALED BY 3D RECONSTRUCTION: IMPLICATIONS FOR AIR-BREATHING EVOLUTION

Takayuki Onai
University of Fukui, Japan

ABSTRACT
Background: Air-breathing has evolved repeatedly in teleosts and often involves remodeling of the cranio-pharyngeal region. In anabantoids such as Macropodus opercularis, the labyrinth organ (LO) develops after metamorphosis from pharyngeal-arch derivatives, and its growth is closely associated with the suprabranchial cavity (SBC), an air-holding space located dorsally to the pharynx. Objective: To identify developmental prerequisites that may facilitate the evolution of SBC/LO-like traits, we analyzed cranio-pharyngeal development in a Cab lineage of Oryzias. Approach: Using 3D reconstruction of post-hatching larvae, we focused on morphological transitions of the epibranchial cartilage and the dorsal pharyngeal roof, which have been implicated in LO-associated morphogenesis. Results: We detected a transient cavity dorsal to the pharynx in ~4 mm total length (TL) larvae (n = 3). This space extended from the posterior end of the eye to just anterior to the otic vesicle. Notably, the cavity was no longer detectable in ~5 mm TL larvae, indicating rapid and stage-dependent remodeling of the dorsal pharyngeal region. Conclusion: These observations suggest that transient formation (and subsequent loss) of a dorsal pharyngeal space is part of normal cranio-pharyngeal development in this Oryzias lineage. Comparative analysis of such spatial dynamics provides a framework to infer developmental conditions required for the establishment and maintenance of SBC-like spaces, offering insights into the evolutionary emergence of air-breathing structures such as the LO.

KEYWORDS
cranio-pharyngeal development; 3D reconstruction; suprabranchial cavity; medaka (Oryzias); air-breathing evolution

HFSP Reference Number: RGP024/2025
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HFSP Award Year: 2025

OSMOTIC POTENTIAL DRIVES LUMENOGENESIS DURING EMBRYONIC MOUSE PANCREAS AND STOMACH DEVELOPMENT

Augusto Ortega Granillo
Max Planck Institute of Molecular Cell Biology and Genetics/Physics of Life Excellence Cluster TU Dresden

Otger Campàs; Anne Grapin-Botton
Max Planck Institute of Molecular Cell Biology and Genetics/Physics of Life Excellence Cluster TU Dresden

ABSTRACT
During embryonic development many organs create hollow cavities, also known as lumens that are essential for physiology. The digestive system comprises several organs with lumens that use them to maintain homeostasis. The stomach has a large single lumen that enables temporary storage and begins food breakdown. In contrast, the pancreas has a tree-like luminal network that collects and empties pancreatic juice near the stomach end. During embryogenesis, both stomach and pancreas develop from posterior foregut endoderm where complex WNT, Bmp, and Fgf signaling controls differentiation of tissue-specific progenitors. While chemical signaling is better understood, the role of physical cues such as osmotic potential is largely unexplored.
In this project we ask how embryonic tissue-specific progenitors modify their physical extracellular environment to achieve functional lumen morphology. Using in vitro organoid systems that give rise to single layer spherical organoids, we study the interplay between organoid growth, lumen osmotic potential, and cell mechanics in the control of tissue-specific growth dynamics. We observed that while lumen formation seems to be similar between pancreas and stomach organoids, after a certain size threshold the organoid volume grows twice as fast in stomach compared to pancreas spheres. Since the lumen represents most of the organoid volume, and osmolyte secretion is thought to drive lumen growth, we measured the osmotic potential inside the lumen. To do this, we injected double-emulsion droplet sensors that can detect osmolarity changes in the lumen. We found that pancreas builds an osmotic potential of 50 kPa across the single cell layer epithelium, at this time measurements in stomach organoids are ongoing. Complementary to these measurements, we conducted acute perturbations of osmolarity in the medium around the pancreas and stomach spheres. We found tissue-specific responses of organoid growth to hypotonic stress. In the long term, we wish to understand the osmotic feedback control that enable different tissues to modulate growth and morphogenesis, as well as identifying the molecular players involved in this feedback mechanism. This work was funded by the Max Planck Society, the Deutsche Forschungsgemeinschaft, and the HFSP organization.

KEYWORDS
Osmotic pressure, embryonic development, lumen formation, mouse development

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HARMONY IN DYNAMICS - DEVELOPING A DIGITAL MULTIPLAYER PLATFORM TO STUDY EMERGENT COOPERATION IN PARTICIPATORY ART

Luciano Perondi
Iuav University of Venice, Italy

Hasan Ayaz[1]; Goni Shifron[2]; Uri Hershberg[3]
[1]Drexel University; [2]Multi-disciplinary independent artist; [3]University of Haifa

ABSTRACT
Emergent cooperation in human groups is difficult to study systematically because naturalistic interaction is challenging to reproduce under scalable and experimentally controlled conditions. The participatory artwork Tä by Goni Shifron, offers a promising paradigm for observing such dynamics: participants manipulate shared pebble arrangements according to simple rules, without verbal communication or explicit coordination, and their initially independent actions gradually organize into synchronized collective behavior. However, the in-person format limits repeated sampling, broad recruitment, and experimental manipulation. To overcome these constraints, we are developing a digital multiplayer version of Tä that enables participants to engage remotely in a shared virtual environment. The goal is to preserve the core interaction principles of the original artwork while creating a platform that supports scalable experimentation on the emergence of cooperation. This raises a central scientific and design question: which elements of the live participatory system are essential for generating emergent collective dynamics, and which can be simplified without altering those dynamics fundamentally? The platform is being developed at the intersection of participatory art, interaction design, multiplayer game design, and behavioral science. Its digital implementation enables systematic manipulation of social and environmental variables, repeated testing across larger and more diverse samples, and more controlled comparison of interaction conditions than is possible in live performance contexts. To evaluate correspondence between the virtual and in-person paradigms, we use validated pre- and post-session measures of affect, anxiety, workload, and social connectedness, including the International Positive and Negative Affect Short Form (I-PANAS-SF), State-Trait Anxiety Inventory- 6 item short form (STAI-6), NASA Task Load Index (NASA-TLX), and inclusion of other in the self (IOS), together with structured observational coding. Ongoing analyses assess whether the digital platform reproduces key behavioral and experiential features of the original Tä interactions. We present the conceptual rationale, design framework, and initial evaluation strategy for the digital Tä platform as a scalable methodology for investigating emergent cooperation. This work provides a foundation for future experimental studies of collective behavior in remotely deployable, systematically manipulable social environments.

KEYWORDS
self organization, synchrnoization, learning, emergant group formation, complex systems

HFSP Reference Number: RGP009/2025
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HFSP Award Year: 2025

UNRAVELING THE LAYERS OF COMPLEXITY UNDERLYING BACTERIA-PLASMID INTERACTIONS

Shai Pilosof
Ben Gurion University of the Negev, Israel

Ying Jie Wang[1]; Kaitlin Schaal[2]; Johannes Nauta[3]; Manlio De Domenico[3]; James Hall[2]
[1]Ben Gurion University of the Negev; [2]University of Liverpool; [3]University of Padova, Italy

ABSTRACT
Plasmids are central drivers of microbial evolution, enabling the rapid spread of traits such as antibiotic resistance between bacteria. Given their critical role in shaping microbe evolution, ecosystem dynamics, and human health, plasmid transmission and maintenance have been intensively studied. Nevertheless, we still cannot reliably predict their dynamics, even in simple microbial communities, because plasmid behavior does not arise from a single process. Instead, it emerges from the interplay between multiple interaction layers operating at different biological scales, which have largely been studied in isolation and without explicit consideration of network structure. In this work, we conceptualize three interconnected network layers that jointly shape bacteria–plasmid community dynamics: (i) the host–plasmid infection network, defining which plasmids can establish within which bacterial hosts; (ii) the plasmid–plasmid compatibility network, determining which plasmids can co-reside within the same cell; and (iii) the horizontal gene transfer network, describing who donates plasmids to whom. Our interdisciplinary team integrates microbiology (J. P. J. Hall; https://www.jpjhall.net/lab), network science (M. De Domenico; https://manliodedomenico.com), and ecological theory (S. Pilosof; https://ecomplab.com) to develop mechanistic models that explicitly embed these interaction structures and test them experimentally. Using stochastic agent-based modeling, we systematically vary network architectures and quantify their independent and joint effects on plasmid persistence, host coexistence, and community stability. Our results demonstrate that infection and compatibility structures generate distinct and interacting dynamical outcomes. In unstructured networks, the community collapses to a single surviving host. Introducing structure in the plasmid–plasmid compatibility network limits highly costly multi-plasmid infections and substantially prolongs transient host coexistence. Structured infection networks increase plasmid diversity and can stabilize host coexistence by partitioning infection niches. Importantly, when re-parameterizing the model to reflect experimentally observed host–plasmid communities, we find that infection network structure alone strongly shapes plasmid prevalence, even in the presence of substantial biological heterogeneity. These experimental results confirm that network architecture is a measurable determinant of community-level plasmid dynamics. Complementing this framework, we further examine how horizontal gene transfer network topology affects plasmid persistence across ecological contexts, showing that large-scale organization governs long-term maintenance and shifts the identity of strains acting as reservoirs under stress.

Together, our findings demonstrate that bacteria–plasmid dynamics are emergent properties of interacting ecological networks. By linking microbiological mechanisms, ecological dynamics, and multilayer network theory, this work provides a general framework for understanding how genetic elements spread and persist in complex microbial communities.

KEYWORDS
ecology, plasmids, microbes, complexity, networks

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HFSP Award Year: 2022

HUMPBACK WHALE SONG RHYTHMS MATCH A MUSICAL UNIVERSAL

Teresa Raimondi
Sapienza University of Rome, Italy
Andrea Ravignani[1]; Peter M. Madsen[2]; Stephanie L. King[3]; Peter F. Cook[4]; Kate Sprogis[5]; Simone Videsen[6]
[1]Sapienza University of Rome; [2]Aarhus University; [3]University of Bristol; [4]New College of Florida; [5]The University of Western Australia; [6]University of Aarhus

ABSTRACT
A comparative approach provides a vital framework for tracing the evolutionary trajectory of musical rhythm. When humans produce music, they conform to universal rhythmic features that transcend cultural boundaries. These 'musical universals' are of profound interest since they might reflect underlying biological mechanisms and functions that are shared, in part, with other mammals. One musical universal is the production of rhythmic categories that coincide with small-integer ratios, i.e., when the durations between successive note onsets are linked by simple relationships (e.g., 1:3, 1:2, 1:1,...). Recent evidence has shown that non-human mammals exhibit a similar rhythmic architecture in their songs. However, mammals that sing and, like humans, show vocal production learning abilities, i.e., the ability to modify vocalizations through social interaction, are rare. Such ability is thought to be crucial in the evolution of human rhythmic capacities. However, one group of mammals exhibits both singing and vocal production learning: whales. In this study, we recorded the songs of four male (escort) humpback whales (Megaptera novaeangliae). These were tagged in 2014, 2018, and 2019 in Exmouth Gulf, Western Australia, with suction-cup tags that record both sound and movement. Humpback whale songs are characterized by a nested structure: the shortest sound is called a unit, and a set of units is combined to form a phrase. Similar phrases are repeated to form a theme, and the combination of themes forms a whole song. First, we automatically annotated the units that compose the phrases, and manually annotated the phrases that compose the themes within a song. Then, we extracted the inter-onset intervals, i.e., the duration between the onset of one event and the onset of the following event, both between adjacent units and between adjacent phrases. At both these (nested) levels of the song, we quantified the rhythmic ratios. i.e., the ratio between adjacent intervals. Their clustering around small-integer ratios was then statistically tested by comparing the number of observed ratios falling in the vicinity of small integers with those far from them. Our findings show that both at the unit- and phrase- level, the rhythmic repertoire matches a crucial universal human musicality, the production of small-integer ratios. Specifically, an isochronous structure (1:1), like the beat of a metronome, dominates the temporal organization between phrases. More complex emerging rhythms (1:2, 2:1), as well as additional rhythmic categories around more complex ratios, were found at the hierarchical level of units. These results contribute to our understanding of vocal rhythm evolution, demonstrating that species sharing key ethological traits with humans also exhibit a comparable rhythmic architecture when singing.

KEYWORDS
musicality, evolution of rhythm, animal songs

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HFSP Award Year: 2022

THE SOCIAL ORIGINS OF RHYTHM: FROM COGNITIVE NEUROSCIENCE TO MARINE MAMMAL FIELDWORK

Andrea Ravignani
Sapienza University of Rome, Italy
Peter Cook; Peter Madsen; Stephanie King
Sapienza University of Rome

ABSTRACT
The capacity to process and entrain to musical beats is a fundamental driver of human social cohesion. However, the selective pressures that originally drove the evolution of social rhythm remain poorly understood. To build testable hypotheses regarding the origins of human rhythmic behavior, this project integrates field biology, comparative neuroscience, bioacoustics, and computational methods. We use marine mammals, which exhibit diverse vocal flexibility but face varying social pressures, as a comparative test-bench to understand broader evolutionary processes leading to communicative rhythms. To robustly quantify these behaviors, we developed metrics and computational tools to measure rhythmic structure in animal vocalizations; in particular, we can now accurately measure how metronomically regular (i.e. isochronous) vocal sequences are, and how precisely and accurately they fall on this metronomic pattern. We also developed agent-based models to test how simulated animals interact vocally in social groups and modify the rhythmicity of their calls based on evolutionary pressures for conspicuousness. Applying these approaches revealed distinct evolutionary applications of rhythm. In wild sperm whales, male slow clicks are highly rhythmic at 0.1 to 0.3 Hz, an order of magnitude slower than other rhythmic signals among mammals and birds. This low-entropy signal may maintain timing information over extreme distances, offering an honest advertisement to distant receivers, with isochrony potentially maintained via extrinsic environmental echoes. Additionally, behavioral tests showed a California sea lion synchronizing movements to complex rhythmic patterns with human-like proficiency. Post-mortem diffusion MRI across three pinniped species revealed brain adaptations directly linking the vocal motor cortex to brainstem vocal motor output nuclei, bypassing midbrain regions that maintain contextual control in most species. This suggests precise temporal control of rhythmic behavior is widely distributed in marine mammals and not dependent on adaptations for social coordination. Conversely, drone observations and playbacks with wild dolphins showed allied males using synchronous motor and vocal displays during cooperative tasks like female herding. Synchrony was more precise among males with weaker social bonds. Playbacks confirmed this synchrony acts as an intrinsic social bonding mechanism for the vocalizing animals. Together, these findings demonstrate that precise temporal control is widely distributed among marine mammals but paint a multifactorial mosaic of rhythmic origins. Crucially, while some aspects of rhythmic behavior are heavily modulated by sociality and cooperative bonding, other rhythmic traits appear completely independent of social coordination. Our work provides comparative data and a methodological-theoretical toolking for understanding the diverse evolutionary roots of social rhythm.

KEYWORDS
cognitive neuroscience, bioacoustics, marine mammal, artificial intelligence

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HFSP Award Year: 2022

CONFINING CHAOS: HOW THE FLOW OF ENERGY ORGANIZES ACTIVE SYSTEMS

Michael Riedl
TU Dresden, Germany

ABSTRACT
All motile active systems, from migrating animal groups to self-propelled synthetic particles, consume energy to sustain motion. In systems with many interacting degrees of freedom, continuous driving readily leads to chaotic dynamics. We find that cytoplasmic flows are likewise inherently unstable: when key organizing mechanisms are perturbed, the flow loses spatial coherence and transitions into chaos. Yet during development, cytoplasmic dynamics remain well coordinated, indicating that mechanisms exist to maintain ordered behavior despite persistent activity.
We show that the cytoplasm actively maintains order by confining its own dynamics in both space and time. Microtubules assemble into asters that generate localized flow domains, thereby reducing characteristic spatial length scales. Simultaneously, the cell-cycle oscillator periodically reorganizes the cytoskeleton, limiting temporal length scales. Through this self-generated spatiotemporal confinement, the cytoplasm reduces the number of accessible dynamical degrees of freedom and prevents sustained chaotic dynamics. If stabilization through confinement restricts accessible dynamical states, it should also reshape how injected energy is processed. To probe this relationship, we developed a minimal synthetic collective of motorized spheres that permits complete, time-resolved measurements of energy input and dissipation at the level of individual agents. In this system, the self-organized transition from disordered to ordered motion is accompanied by a redistribution of dissipation from internal losses to coordinated external mechanical work. We show that the transition to order is accompanied by a measurable redistribution of dissipation that alone distinguishes dynamical states. Our findings indicate that order in active systems emerges when energy dissipation reorganizes from numerous uncorrelated pathways into coordinated modes. Developmental robustness thus reflects the cytoplasm's ability to sustain this energetic organization.

KEYWORDS
cytoplasmic flows, active matter, self-organization, chaos, energetics

HFSP Reference Number: CDF0049/2024
HFSP Award Category: Cross Disciplinary Fellowship
HFSP Award Year: 2024

UNIVERSAL BIOCHEMISTRY OF RNA IN THE COLD

Felix Ritort
Universitat de Barcelona (UB), Spain

ABSTRACT
RNA impacts biological diversity and life. The main difference with DNA is the presence of the ribose 2'OH hydroxyl group, which enables RNA to hydrogen-bond with nucleotides and water and to catalyze reactions mediated by metal ions. Recent single-RNA pulling experiments in our lab with a temperature-jump optical trap in the temperature range 5-50ºC found that RNA hairpins invariably form alternative misfolded structures below a glass transition temperature TG = 20ºC where the heat capacity of the folded hairpin abruptly drops. Moreover, maximum stability is found at TS= 5ºC where the water density is maximum, and cold denaturation transition is predicted to occur at TC= -50ºC. The prominent role of the ribose-water clathrate in the cold suggests that TG, TS and TC are universal temperatures weakly modulated by sequence. The promiscuous sequence-independent ribose-water interactions mediated by the 2'OH hydroxyl group portend an altered RNA biochemistry at low temperatures, with consequences for novel RNA functionalities and catalytic activities at freezing and subfreezing temperatures, for RNA function in present-day cold-loving psychrophilic organisms, and potentially for RNA evolution.

KEYWORDS
RNA folding, single molecule experiments, force spectroscopy

HFSP Reference Number: RGP017/2025
HFSP Award Category: Research Grant
HFSP Award Year: 2025

WHY DON'T WE GET MORE CANCER? A TISSUE-LEVEL PARADOX IN ONCOGENIC CELL FATE

Felipe Rodrigues
The Rockefeller University, USA

ABSTRACT
Every human accumulates millions of cells carrying cancer-driver mutations over a lifetime, yet most of these mutant cells never progress to cancer. The skin — our primary barrier to the outside world — is constantly exposed to environmental insults including UV radiation, wounding, and pathogens. These triggers can induce mutations, inflammation, or both, creating a tissue environment in which mutant cells and inflammatory signals coexist. Remarkably, studies of normal human skin reveal that by middle age, the epidermis is extensively colonised by positively selected cancer-driver mutations. What prevents these mutant cells from expanding and initiating malignancy? A leading hypothesis implicates inflammation as a promoter of tumour development, but the relationship between inflammation and the fate of mutant cells in living tissue remains poorly understood. Using the mouse skin epidermis as a model, we track the dynamics of cells carrying an oncogenic mutation (HRASG12V) prevalent in human skin cancers, in an otherwise normal tissue. Paradoxically, we find that multiple distinct inflammatory stimuli, applied acutely or chronically, consistently reduce the abundance of mutant epithelial cells relative to their wild-type neighbours. This suppressive effect persists across multiple genetic contexts and is not specific to HRAS, suggesting a general tissue-level response that limits the persistence of oncogenic cells during inflammation. Yet inflammation is not simply protective: mice with a prior history of epidermal inflammation show elevated tumour burden and more invasive pathology following chemical carcinogenesis. This creates a striking paradox: inflammation simultaneously reduces the abundance of mutant cells and primes the tissue for malignancy. We propose that inflammation remodels the cellular landscape of the epithelium. It suppresses mutant cells in the short term, but enriches for the rare clones capable of enduring this pressure, potentially the very cells that go on to form tumours. Understanding this dynamic may help explain why cells with oncogenic mutations do not always give rise to cancer — and what tips the balance when they do.

KEYWORDS
Cancer initiation, Inflammation, Stem cells, Oncogenic mutation, Tissue homeostasis

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HFSP Award Category: Long-Term-Fellowship
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DARK OXYGEN PRODUCTION: AN OVERLOOKED PROCESS IN EARTH'S HIDDEN ECOSYSTEMS

S. Emil Ruff
The Marine Biological Laboratory, USA

Jordon D. Hemingway[2]; Emeline Vidal[1]; Carolina Carvalho[2]; Tetyana Gilevska[3]; Toshiki Nagakura[3]; Laura Schwab[4]; Ranjani Murali[5]; Beate Kraft[3]
[1]The Marine Biological Laboratory; [2]ETH Zürich; [3]University of Southern Denmark; [4]University of Jena; [5]University of Nevada Las Vegas

ABSTRACT
Nearly all molecular oxygen (O2) on Earth is produced in sunlit environments via photosynthesis by plants or microorganisms. Consequently, the light-independent or "dark" O2 production has been thought to be negligible. Permanently dark environments in Earth's subsurface, such as oil reservoirs, groundwater aquifers and seafloor sediments were thus historically presumed to function under conditions largely devoid of O2. Contrary to this, with support of HFSP, we are showing that O2 is produced and consumed in dark and apparently anoxic environments at a much larger scale than previously assumed. Dark oxygen production occurs both abiotically, e.g. through physical and chemical reactions in rocks and water [1], or through specialized microbial metabolisms, which involve the dismutation of nitric oxide or chlorite [2]. Our team has shown that microbes and metabolisms capable of dark O2 production are globally widespread and abundant in diverse subsurface ecosystems, for example groundwaters [3, 4]. This enables obligate aerobic (i.e., O2-dependent) microorganisms to thrive in many environments that were deemed unsuitable for aerobic life. Our genomic analyses reveal that the capacity for dark O2 production is far more widespread across the tree of life than previously recognized. For example, one of Earth's most abundant microbial clades, Nitrosopumilus maritimus, can produce O2 in the dark [5]. Beyond genomic inferences, our analysis of oxygen isotopes of O2 in groundwater provides chemical evidence to suggest dark O2 production in up to half of the studied aquifer environments [2]. This observation improves our understanding of the functioning of groundwater ecosystems, with potential relevance for humanities' global use of groundwater as a source of drinking water. In summary, our microbiological and geochemical data support the conclusion that dark O2 production is an important and widespread process in Earth's subsurface environments. The gained insights challenge long held assumptions in subsurface biogeochemistry and ecology and are critical for an improved understanding of global subsurface element cycles. Widespread dark O2 production also carries far-reaching implications concerning the sources, fluxes and effects of O2 on early Earth environments and potentially on other celestial bodies.

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KEYWORDS
subsurface microbiome, oxygen cycling, deep life, dark oxygen production, microbial ecology

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WHEN THE WEAKER TEAM OUTPERFORMS THE STRONGER ONE: A REINFORCEMENT LEARNING AND COMPLEX SYSTEMS ANALYSIS

Miguel Ruiz-Garcia
Complutense University of Madrid, Spain

Rachael Skye[1]; Samuel Lozano Iglesias[2]; Samuel Liebana[3]; Younes Strittmatter[4]; Andrew Saxe[3]; Markus Spitzer[6]; Erin Teich[1]; Miguel Ruiz-Garcia[2]
[1]Wellesley College; [2]Complutense University of Madrid; [3]University College London; [4]Princeton University; [6]Martin Luther University Halle-Wittenberg

ABSTRACT
Collective behavior is a key component of humanity's success story. Through interaction and collaboration, groups of humans have built bridges, discovered vaccines, developed complex languages, and accumulated over generations a collective knowledge stored in the structure of society. In collaborating groups, collective intelligence describes the capacity of groups to achieve levels of performance that cannot be explained by the abilities of individuals alone. In particular, group composition, including the relative skill level of team members, can alter collective behavior and may either suppress or enhance group performance. To study this problem in a controlled environment, we have designed collaborative multiplayer online games that allow us to manipulate team makeup and coordination demands by varying player abilities and their environment. We explore how collective intelligence is encouraged or reduced by constraints in the environment and how it is shaped by the relative abilities of team members. Remarkably, we find that under specific conditions, teams of players with higher average ability can be outperformed by lower average ability teams with complementary strengths. To understand these effects, we analyze team behavior using methods from complex systems, combined with reinforcement learning simulations and simplified dynamical systems models amenable to analytical descriptions. These approaches together help us to understand the key factors controlling collective behavior.

KEYWORDS
collective intelligence, reinforcement learning, complex systems

HFSP Reference Number: RGEC33/2024
HFSP Award Category: Research Grant Early Career
HFSP Award Year: 2024

INTRACELLULAR VOLTAGE CONTROL OF DECISION-MAKING DURING CELL MIGRATION

Pablo J. Sáez
University Medical Center Hamburg- Eppendorf | UKE, Germany

Yamuna Krishnan[1]; Nir Gov[2]
[1]The University of Chicago; [2]Weizmann Institute of Science

ABSTRACT
Cell migration is pivotal for wound healing, immunity, and cancer progression. As cells move through tissues, they navigate complex microenvironments, encountering barriers and neighboring cells. During directional migration, chemical and physical cues provide a bias that guides cells along preferred paths, where obstacles generate branching arms and one ultimately becomes the chosen direction. However, much less is known about how cells decide for a new direction in absence of bias. Cells sense microenvironmental cues partly through ion channel-driven changes in membrane potential (MP), yet the connection between MP and directional choice has remained unclear. Organelle ion channels also influence MP, and migrating cells use polarity as a navigation system, which in turn is reflected in organelle organization, raising key questions. How do single cells choose a direction of migration? Do changes in MP regulate this decision? Here, we combined cell biology, biological chemistry and new theoretical frameworks to address these questions. We developed new molecular tools and used live-cell imaging to validate a new mathematical model to explain how cells choose a new direction. We created high-resolution reporters for ions and pH, including pHlicKer, which revealed that cell-surface K^+ channels are active in organelles, and RatiNa, which showed that lysosomes help cells withstand extracellular salt stress. Using our DNA-based, pH-correctable Ca^{2+} reporter CalipHluor, we identified TMEM165 as the first lysosomal Ca^{2+} importer. With SeRapHin, a chemigenetic pH reporter, we uncovered pH gradients across major membranous organelles, including the ER and mitochondria. Interestingly, mitochondrial MP oscillations at the leading edge of motile cells correlated with directional decision-making, suggesting that mitochondrial function steers migration. To understand how cell direction is determined, we used micropatterns to force cellular branching during migration, and developed machine learning-based image analysis to track branch behavior. In single junctions (Y-shaped) actin-based membrane oscillations allowed cells to explore both options until one branch exhibits stronger retrograde flow, subsequently defining the winning arm. Cancer cells were faster to decide in comparison to non-cancerous cells. In highly branched cells, similar oscillations occurred, along with a trade-off between branching and cellular polarity: branching provided greater exploration, but it reduced overall migratory efficiency as cells became trapped. Immune cells showed a higher threshold for this trade-off and were able to migrate despite high branching. Our coarse-grained model predicted these behaviors and extended to leukocyte migration in vivo (zebrafish), where polarity and shape dynamics balanced exploration (branching) and migration (speed).

Together, our experiments and our new theoretical framework describe and predict how cells choose migration paths. Indeed, the ability to predict cell migration is an essential precursor to controlling its movement to accelerated wound healing, prevention of metastasis or sculpting of the immune response.

KEYWORDS
decision-making, membrane potential, cell biology, chemistry, modelling

HFSP Reference Number: RGP0032/2022
HFSP Award Category: Research Grant Program
HFSP Award Year: 2022

LOCAL EXTRACELLULAR MATRIX REMODELING AND NEURON-GLIA SIGNALING PROMOTE BRAIN PLASTICITY AFTER INJURY IN DROSOPHILA

Juan Andrés Sánchez
Champalimaud Foundation, Portugal

Mariana B. Santos[1]; Carolina Alves[1]; Anabel Rodriguez Simões[1]; Ana Cristina Ojalvo-Sanz[1]; Sara Monteiro Ferreira[1]; Patricia Borges[1]; Adriana Sánchez Danés[1]; Sergio Casas Tintó[2]; Christa Rhiner[1]
[1]Champalimaud Foundation; [2]Carlos III Health Institute

ABSTRACT
The adult optic lobes of flies serve as a powerful model system for investigating the intricate mechanisms of adult brain plasticity, particularly in response to physical damage. Within this context, a population of normally quiescent cells possesses the remarkable ability to exit their dormant state and undergo active proliferation. In this study, we delve deeper into the molecular underpinnings of this regenerative process by examining transcriptional change specifically in sorted proliferating cells marked by mitotic-dependent labelling. Among the earliest and most strongly upregulated factors, we identified a Heparanase-like protein predicted to bind and restructure the local extracellular microenvironment. Functional analyses demonstrate that this extracellular matrix (ECM) remodeling factor promotes repair and animal survival following injury. Consistent with a central role for ECM dynamics, we detected rapid local injury-induced ECM alterations in Heparan Sulfate Proteoglycans (HSPGs) within the optic lobe. Notably, the glypicans Dally and Dlp exhibit distinct and spatially restricted expression changes after damage, suggesting specialized roles in brain repair. Mechanistic studies further reveal that glypicans and glypicans regulators, such as the secreted Heparanase-like protein, regulate Dpp (the TGF-β/BMP orthologue) signaling. This Dpp-dependent neuron–glia communication is required to activate regenerative plasticity in the adult brain. Notably, we found that Heparanase-like proteins are also active in highly malignant brain tumors such as glioblastoma in the brain of flies and medulloblastomas in mice, highlighting a shared mechanism between regeneration and tumor progression. Together, our findings reveal a dual role for ECM dynamics in both tissue repair and cancer progression, offering new insights into the molecular basis of brain plasticity.

KEYWORDS
Brain injury, Extracellular matrix remodeling, TGF-Beta/BMP signaling, Brain tumors, Drosophila

HFSP Reference Number: LT0014/2024-L
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HFSP Award Year: 2024

DECODING ENDOMETRIAL COMPARTMENTALIZATION WITH MOUSE UTERINE EXPLANTS

Fatima Sanchis Calleja
Harvard University, USA

Kara Mckinley
Harvard University

ABSTRACT
The inner lining of the uterus (endometrium) is the most powerful regenerative system in the human body. It sheds and regenerates with each menstrual cycle, repeating this process up to 400 times over the human reproductive lifespan. Nearly one quarter of the world's population menstruates, and hundreds of millions suffer from menstruation-associated disorders. Yet menstruation remains poorly understood due to social stigma, low funding, and the scarcity of animal species that naturally menstruate. Every month, the endometrium prepares for pregnancy through a one-way differentiation process called decidualization. In the absence of pregnancy, decidualized tissue is shed during menstruation and then rebuilt from remaining endometrial tissue. Traditionally, it is thought that the upper layer of the endometrium (functionalis) is lost, while the deeper layer (basalis) remains and regenerates the functionalis. This basal-luminal compartmentalization is crucial: damage to the basalis is associated with non-regenerative outcomes (fibrosis). Using an inducible model of menstruation in mice, recent work from our group revised this model, showing spatiotemporal regulation of decidualization and shedding of distinct endometrial regions in a radial-like pattern. However, the origin of those patterns remains unknown. Existing human organoid models lack key cell types involved in menstruation (e.g. immune cells) as well as a stereotypical in vivo-like morphology. In mouse models, the spatial orientation of the uterus makes the endometrium inaccessible to intravital imaging techniques. Here, we developed and optimized a novel system to study endometrial compartmentalization ex vivo using mouse uterine explants - cross-sections of fresh uteri that preserve the full basal-to-luminal axis and native tissue architecture. This platform enables precise spatiotemporal manipulations of hormonal and molecular signals as well as longitudinal imaging in a genetically tractable model. Together, these features make a powerful tool to understand endometrial compartmentalization and its role in decidualization, menstrual shedding, and regeneration, ultimately informing future therapeutic strategies for menstrual disorders.

KEYWORDS
Menstruation, decidualization, transgenic mouse models, live imaging, explants

HFSP Reference Number: LT0039/2024-L
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MODELING AND EXPERIMENTAL APPROACHES ALLOW US TO PARSE DRIVERS OF COMMUNITY RESPONSES TO ENVIRONMENTAL CHANGE

Kaitlin A. Schaal
University of Liverpool, UK

Ying-Jie Wang[2]; Johannes Nauta[3]; Manlio De Domenico[3]; Shai Pilosof[2]; James P. J. Hall[1]
[1]University of Liverpool; [2]Ben-Gurion University of the Negev; [3]University of Padua

ABSTRACT
Microbial communities perform key roles within global ecosystems, as their members interact to carry out metabolic functions. Their composition is shaped by interactions, such as competition and horizontal gene transfer, where members may eradicate one another or share plasmids that enable the spread of metabolic or antibiotic resistance genes. These are qualitatively different interactions that occur at the same time, making it difficult to untangle their relative contributions to overall community dynamics. Our research team studies interaction dynamics of plasmids and the bacteria which host them over short (ecological) and long (evolutionary) time scales in order to understand the key drivers necessary for predicting outcomes. We focus on three types of microbial community interactions: plasmid-plasmid interactions (such as incompatibility and gene transfer), bacteria-plasmid interactions (such as host range and the fitness effects plasmid have on their hosts), and bacteria-bacteria interactions (such as interspecies competition and cooperation). Considering each interaction type as a network allows us to study how the outcomes of interactions in one network affect dynamics in the other networks. We have used models and experiments to disentangle complex interactions in bacteria-plasmid communities and understand responses to environmental change. We have developed a laboratory model system of three bacterial species and two plasmids, where each plasmid can be hosted by two species and one species can host both plasmids. We have previously shown that species interactions influence plasmid maintenance, even under environmental conditions where plasmids are beneficial, as bacterial competition is influenced by species-specific costs of environmental stress and plasmid burden. Here we take advantage of temperature-dependent growth rates to modulate the bacteria-bacteria interaction network and explore how species competitiveness affects community dynamics and plasmid-borne resistance in the face of abiotic perturbations. We show that altering the relative competitiveness of the three bacteria changes the community response to environmental perturbations. Host and plasmid dynamics influence each other - in some cases, a plasmid can hitchhike to high frequency within a host population even when the environmental conditions select for the other plasmid. Together with an agent-based model, we show that relative competitiveness of the three species determines community response to environmental stress, with plasmid cost and loss (segregation) in turn shaping bacterial population dynamics. Overall, we show the power of our combined approach, using experiments and models, to provide insight into complex bacterial community dynamics. As anthropogenic environmental perturbations continue to impact ecosystems, understanding the factors that shape microbial communities becomes increasingly important for predicting and managing ecosystem resilience.

KEYWORDS
plasmids, community interactions, microbial ecology, environmental change

HFSP Reference Number: RGY0064/2022
HFSP Award Category: Research Grant Early Career
HFSP Award Year: 2022

BLOOM-IN-A-LAB: SELF-ORGANISED ACTIVE TRANSPORT MITIGATES NUTRIENT CONSTRAINTS IN ALGAL BLOOMS

Anupam Sengupta
Luxembourg University, Luxembourg

ABSTRACT
As the ocean warms and becomes more strongly stratified, blooms of toxin-producing phytoplankton are expected to encounter a basic physical limitation: sharp density gradients suppress vertical mixing, confining cells to thin layers where light and nutrients seldom coincide. Yet the recurring dominance of motile, gravitactic phytoplankton during harmful algal bloom (HAB) events suggests an alternative mechanism—collective, self-generated flows that reorganize resource distributions from within the bloom itself. Using our Bloom-In-A-Lab platform, we uncover emergent fluid motions within blooms and, their mechanistic role in conferring resilience to blooms under resource limitations. Combining quantitative imaging with deep-learning–based tracking and segmentation, we directly observe bioconvective plume formation and measure how these plumes redistribute dissolved nutrients and microscopic cargo when both light and nutrients are scarce. Our results show that plume dynamics are not simply a byproduct of motility, rather distinct cellular phenotypes actively regulate the timing, spatial organization, and turnover of plumes, thereby modulating transport efficiency. This control supports energy-efficient migration and reduces disparities in resource access across the population, effectively linking individual traits to community-scale “pumping” of the surrounding fluid. In mixed communities comprising multiple phenotypes and species, these trait differences correspond to distinct flow-coupled pathways, producing spatiotemporally segregated structures rather than a single, homogenized bloom state. Together, these findings indicate that microbial self-organization can act as an intrinsic driver of HAB persistence, particularly as climate change intensifies ocean stratification.

KEYWORDS
gravitaxis, active flow, nutrient uptake, algal bloom

HFSP Reference Number: LT000993/2014
HFSP Award Category: Cross Disciplinary Fellowship
HFSP Award Year: 2014

CALCIUM-RESPONSIVE LIPOSOMAL NANOPARTICLE REPORTERS FOR AMPLIFIED MOLECULAR MRI OF EXTRACELLULAR CALCIUM

Vinay K Sharma
Massachusetts Institute of Technology (MIT), USA

ABSTRACT
Sensitive detection of neurochemical dynamics by molecular functional MRI remains limited because most paramagnetic sensors produce only modest relaxivity changes upon analyte binding. Liposomal nanoparticle reporters (LisNRs) were previously introduced as an amplified sensing platform in which high concentrations of gadolinium-based contrast agents are encapsulated within lipid vesicles containing analyte-responsive membrane components, enabling signal transduction through changes in water permeability and collective modulation of longitudinal relaxivity [1,2]. Initial validation with a biotin-responsive formulation demonstrated the feasibility of this membrane-based amplification strategy for molecular imaging [2]. Here, calcium-responsive LisNRs (Ca-LisNRs) were designed to enable MRI detection of extracellular Ca^{2+}, a key regulator of neuronal excitability and synaptic signaling. The design objective was to convert calcium recognition into amplified MRI contrast through membrane-level structural changes rather than single-site chelation. Ca-LisNRs incorporate BAPTA-derived lipid switches within the liposomal bilayer while maintaining a high internal payload of gadolinium contrast agent. Calcium coordination induces collective lipid rearrangements that transiently increase membrane water permeability, producing amplified and reversible increases in longitudinal relaxivity at physiologically relevant calcium concentrations with strong ion selectivity. Systematic variation of lipid composition established structure–function relationships governing sensitivity, stability, and reversibility, and identified formulations suitable for in vivo application. Following intracranial delivery in the rat brain, Ca-LisNRs generated EGTA-sensitive and reversible T_1-weighted contrast changes, supporting proof-of-principle detection of extracellular calcium dynamics in living tissue. These results establish calcium-responsive LisNRs as an amplified molecular MRI platform and extend membrane-transduced nanosensor strategies toward functional imaging of biologically relevant ions in vivo.

References:
1. Simon J et al. (2023) Nat Biomed Eng 7: 313–322.
2. Das S et al. (just accepted) Nat Biomed Eng.

KEYWORDS
Liposomal nanosensors, calcium sensors, molecular fMRI, MRI contrast agents

HFSP Reference Number: CDF0027/2024
HFSP Award Category: Cross Disciplinary Fellowship
HFSP Award Year: 2024

UNDERSTANDING THE EVOLUTIONARY SIGNIFICANCE OF PIONEER TRANSCRIPTION FACTORS IN INSECT METAMORPHOSIS

Yu Shirai
Harvard University, USA

Suhrid Ghosh; Cassandra Extavour
Harvard University

ABSTRACT
Insects are the most diverse group of animals on Earth. One of the major factors that has enabled their great success is the evolution of metamorphosis, which partitions their life cycles into juvenile and adult stages. Despite its profound significance, the evolutionary origin of insect metamorphosis remains largely enigmatic. Several transcription factors have been shown to play essential roles in metamorphosis. One of these master regulators is the adult specifier E93, which is required for adult differentiation by modulating chromatin accessibility. E93 transcripts are highly expressed at pre-adult stages in both hemimetabolous (whose juveniles are similar to the adult) and holometabolous (whose juveniles are different from the adult) insects. Intriguingly, in hemimetabolous insects E93 transcripts also exhibit peak expression levels at the early stages of embryogenesis. This suggests an ancestral role of E93 in embryogenesis, and that evolved changes in its expression pattern could be a key evolutionary event enabling the "juvenile-to-adult" transition in insects. Based on the specific expression pattern and known molecular functions, I hypothesize that the ancestral role of E93 is as a maternal-to-zygotic transition (MZT) pioneer factor, driving zygotic genome activation. In this project, I aim to elucidate the ancestral role of E93 and compare it with known MZT pioneer factors (Zelda, GAGA factor, CLAMP), characterized in Drosophila, by exploring their roles in a hemimetabolous model, the cricket Gryllus bimaculatus.

KEYWORDS
insects, metamorphosis, pioneer factor, Evo-Devo

HFSP Reference Number: LT0048/2025-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2025

EMERGENT COLLECTIVE BEHAVIOR IN HUMANS PLAYING ONLINE GAMES

Rachael Skye
Wellesley College, USA

Younes Strittmatter[2]; Erin Teich[1]; Markus Spitzer[3]
[1]Wellesley College; [2]Princeton University; [3]Martin Luther Universität Halle Wittenberg

ABSTRACT
Collaboration and interaction is key to the ability of individuals to work towards larger goals across all scales of life, from ants foraging to the motion of fish schools to human teams. In particular, collaboration does not always require explicit communication: for instance, schools of fish change direction far faster than an explicit signal would be propagated through the group, instead responding to changes in their local environment. While human collective behavior has been investigated across various contexts, a comprehensive theory describing the emergence of this behavior has yet to be developed. We present work that examines collaboration in groups of humans as they play a well-controlled and tunable online game, in which players can work together but cannot explicitly communicate. We examine collaboration through the lens of statistical physics and network theory, and find that collaboration among humans reliably emerges and can be enhanced or suppressed by manipulating the game environment and relative player ability. Our results contribute to a broader understanding of emergent collective intelligence during active team collaboration, and point toward causal mechanisms for that emergence in a model environment.

KEYWORDS
collective intelligence, complex systems, human behavior

HFSP Reference Number: RGEC33/2024
HFSP Award Category: Research Grant Early Career
HFSP Award Year: 2024

A QUANTITATIVE IN VITRO APPROACH TO VIRUS INFECTION

Paul Soudier
University of Minnesota, USA

Antoine Levrier; Vincent Noireaux Paul
University of Minnesota

ABSTRACT
Virus-host interactions are fundamental biological processes that have shaped life through evolution. Their extensive study is crucial for a wide diversity of purposes ranging from the deciphering of fundamental mechanisms governing biological organisms at all scales to the many applications reached in medicine, biotechnologies and other fields. Bacteriophage, a class of bacteria infecting viruses that constitute the most abundant biological entities on earth, have been historically used as models to study virus-host interactions. After their discovery in the 1910s, their study in the mid 20th century has enabled the description of some of their key structures, functions and associated processes while also participating in the shaping of the emerging field of molecular biology. In 1974 Arthur Kornberg noted that "Despite the extraordinary complexity of the viral life cycle, its dissection and eventual reconstruction with chemically defined components in a cell-free system from start to finish remains an attractive prospect" (Kornberg, DNA Replication, 1974). Strikingly, despite our extensive knowledge of phage evolution, ecology, and molecular mechanisms, a complete in vitro phage infection cycle based on a synthetic cell (SC) system has not been achieved, underscoring both conceptual and technical frontiers. Leveraging recent advances in the fields of in vitro transcription translation systems and synthetic cell engineering, we sought to advance toward Kornberg's vision by demonstrating the reproduction of a full lytic phage infection cycle in a controlled in vitro setting. Focussing on the model lytic coliphage T7, we engineered cell-free expression reactions encapsulated in liposome-based synthetic cells displaying lipopolysaccharide that serve as a sole receptor for T7 phage. In this system we were able to reconstitute key steps of a phage infection cycle including phage binding to the SCs via a specific receptor, T7 genome ejection into the SCs, genome replication within the SCs and active phage synthesis and assembly inside the SCs. We quantitatively characterize each step, notably the multiplicity of infection, the replication efficiency, the liposome size constraints on infection rates, and the phage rebinding dynamics. This in vitro reconstituted phage cycle provides a bottom-up approach to studying viral infection in a controlled environment, decoupling phage biology from the complexities of living bacteria.

KEYWORDS
Bacteriophage, synthetic cells, synthetic biology

HFSP Reference Number: LT - 0007/ 2024
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2024

KINETIC PROFILING OF TRANSCRIPTIONAL ACTIVATION DOMAINS WITH TRANSCRIPTIONAL RESPONSE BARCODE LINKED SEQUENCING (TREBL-SEQ)

Max Staller
UC Berkeley, USA

Antoni Wrobel; Marcin Suskiewicz
UC Berkeley

ABSTRACT
Transcription factors regulate the kinetics of gene expression with DNA binding domains and activation domains. DNA binding domains are conserved, structured and bind to specific DNA sequence motifs. Activation domains are poorly conserved, and intrinsically disordered, features that have made them difficult to study with traditional methods. Activation domains recruit coactivator complexes to drive transcription. Transcription factors regulate the kinetics of gene expression through post translational modification that regulate nuclear localization and/or DNA binding. Using a new high-throughput assay, we have found that the protein sequence of activation domains can directly regulate transcriptional kinetics. Some activation domains mediate fast and strong transcriptional responses while others mediate slow but equally strong responses. We identify the protein sequence features that control the speed and strength of these transcriptional responses.

KEYWORDS
transcription, transcription factor, gene expression, dynamics, kinetics, machine learning, neural networks, intrinsically disordered proteins

HFSP Reference Number: RGEC30/2025
HFSP Award Category: Research Grant Early Career
HFSP Award Year: 2025

NEUROGENIC MECHANISMS IN THE LARGEST INVERTEBRATE BRAIN

Jessica Stock
Harvard University, USA

Alyson Hally[2]; Julia Lucey[1]; Loyal Goff[2]; Caroline Albertin[1]
[1]Harvard University; [2]Johns Hopkins University School of Medicine

ABSTRACT
Coleoid cephalopods (octopus, squid, cuttlefish) present a remarkable example of convergent evolution, as well as evolutionary innovation. They have evolved the largest invertebrate brain, entirely independent to the vertebrate lineage. With around 500 million neurons, the nervous system of Octopus vulgaris far exceeds that of its closest relatives (slugs, clams, etc.) and even rivals that of many vertebrates in size and neural complexity. The foundation for the structure and organization of the cephalopod brain is laid during embryonic development; however, its impressive size is largely achieved through post-hatching neurogenic growth, with some species increasing brain mass over 100-fold in size between hatching and adulthood. Contrarily, mammals exhibit very limited post-embryonic neurogenesis, that is located in specific areas embedded within the respective brain regions. Therefore, cephalopods provide a unique opportunity to both, understand fundamental principles of how large nervous systems are built, and explore novel neurogenic processes that drive post-embryonic brain expansion. To identify post-embryonic neurogenic territories and mechanisms that drive incorporation of newly born neurons into the growing brain, we have established a suite of molecular tools, including birth-dating studies, spatial gene expression assays, whole-mount imaging techniques, and tissue grafting approaches, in the hummingbird bobtail squid Eurpymna berryi. Intriguingly, we found no evidence for cell proliferation in the developing brain, but rather that post-hatching neurogenesis takes place outside the central nervous system. We characterize the white body, a tissue located behind the eyes, as the main source for new neurons after embryonic development. To integrate into the central brain complex, newly born neurons undergo extensive, long-range cell migration, while regional identity within the white body dictates their migratory route and final position in the nervous system. This work provides fundamental insights into the formation of the largest invertebrate brain, describes a novel mechanism of post-embryonic neurogenesis that drives extensive brain expansion and starts to elucidate how new neurons are incorporated into existing circuits of the massive cephalopod nervous system. Ultimately, this research has the potential to describe alternative approaches evolved in cephalopods to tackle common neural engineering challenges found in vertebrates and across the animal kingdom.

KEYWORDS
neurogenesis, evolution, development, cephalopod

HFSP Reference Number: LT0038/2023-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2023

DECIPHERING CIRCUIT MECHANISMS OF COGNITION WITH LARVAL ZEBRAFISH

Ryosuke Tanaka
RIKEN Center for Brain Science, Japan

ABSTRACT
An important hallmark of our cognitive abilities is retention of information over time. For example, when you look for your favorite pair of socks from a drawer or when you compare bottles of wine, your brain is representing information that is no longer given to your sensory systems over many seconds. Such sustained mental representations are thought to be supported by continuous firing of neurons. However, it is not trivial how neurons can keep firing for many seconds, given the millisecond-order timescales of individual neurons. Theoretical studies have suggested that recurrent circuit structures can create sustained neural dynamics from fast neurons, yet it remains extremely challenging to map exact circuit structures in large brains of mammals, where neural bases of cognition are typically studied. To overcome this challenge, I focus on the larvae of zebrafish, one of the smallest vertebrate model organisms. The numerical simplicity of their brain with only 100,000 neurons, together with the availability of various transgenic tools to monitor and manipulate activities of specific neuron types, make it a real possibility for us to understand how recurrent circuitry generates sustained neural dynamics and support cognition. In this presentation, I will discuss my recent projects that uncovered (1) how larval zebrafish remember past wide-field visual motion for stabilizing their swimming course in the future (Tanaka & Portugues, 2025; doi: 10.1016/j.cub.2025.08.047), as well as (2) how the neural compass of larval zebrafish integrate various visual cues to improve their head direction estimates (Tanaka & Portugues, 2026; doi: 10.1038/s41586-025-09888-x). I will also introduce my most recent efforts to leverage more advanced genetic tools (e.g., transcriptomics, fluorescent visualization of neurotransmitters) to dissect neural circuits surrounding the fish neural compass.

KEYWORDS
neuroscience, cognition, attractor dynamics, navigation, zebrafish

HFSP Reference Number: LT0027/2023-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2023

IN TUNE WITH THE MOON: THE EVOLUTION OF MOON-CONTROLLED TIMING SYSTEMS FROM THE SURFACE TO THE DEEP SEA

Kristin Tessmar-Raible
AWI (Alfred-Wegener-Institute Helmholtz Centre for Polar and Marine Research), University of Vienna, Austria

Audrey Mat[1]; Federico Scaramuzza[1]; Marjolaine Matabos[2]; Orit Peleg[3]; Todd Oakley[4]
[1]University of Vienna; [2]IFREMER; [3]University of Colorado; [4]University of California, Santa Barbara

ABSTRACT
Since its beginnings, life has been exposed to the regular environmental cycles caused by sun and moon, requiring adjustments of physiology and behavior. We investigate different biological rhythms and their underlying oscillators in marine environments, a subject of relevance for evolution, ecology and climate. In the context of our HFSP project, our work focusses on the ecological key deep-sea mussel, Bathymodiolus azoricus, and its closest surface relatives of the Modiolus genus. For those species, we analyse the different endogenous oscillators and their interaction with environmental cues from population-level to individual physiology and molecular cell biology. We find that even though the deep-sea hot vent environment lacks obvious daily cues, endogenous, circadian oscillators are present on cellular level in the deep-sea Bathymodiolus mussels. These mussels also exhibit tidal rhythms on tissue and behavioral level, whose mechanistic origin remains, however, currently unclear. Albeit solar (and lunar) light is absent at the mussels natural habitat at -1700m, both rhythms can be influenced by light (– see separate abstract by Mat, A et al). We also integrate and compare the data acquired from the mussels with our research in marine annelids (KT-R) and ostracods (TO) at surface level. Both species also exhibit combinations of different rhythms driven by endogenous timers. Functional genetics and modeling in the annelid Platynereis allowed us to uncover a mechanism by which the period length of an endogenous ~24h clock can be tuned to run with different speed depending on the phase of the lunar cycle and lunar light. This plastic circadian-circalunidian oscillator adjusts the exact timing of worms' mating behavior. Of note, albeit functional interference is still lacking for the mussels, their co-occurring circatidal and circadian rhythms are likely also best explained by a certain level of plastic functions of what are commonly known as core circadian clock genes. These "core circadian clock genes" may not be strictly circadian, but rather considered "tunable", depending on the required period length. On the other hand, the worms' monthly (circalunar) oscillator, which is also controlled by the moon, but runs with a much longer period length, does not depend on core circadian clock genes. Overall, our work starts to provide insight into the question how different lunar-controlled timing systems are set up across very different marine environments and evolution. It thereby also emphasizes the concept that oscillator systems and genes commonly considered to be "circadian" can be more plastic, and allows us to carve out the mechanistic similarities and differences of these oscillators in the context of moon-dependent "tuning".

KEYWORDS
chronobiology, marine, ecosystems, molecular biology, modeling

HFSP Reference Number: RGP021/2024
HFSP Award Category: Research Grant Program
HFSP Award Year: 2024

COUPLED CHEMICAL MEMORY AND COMPUTATION AT THE SINGLE-MOLECULE LEVEL

Duhan Toparlak
University of Oxford

ABSTRACT
A single ribosome reads genetic code and manufactures proteins simultaneously, performing memory retrieval and translation, i.e., computation, as one inseparable act. Artificial molecular systems can also store data and chemical reaction networks can implement reservoir computing, but a bottom-up design that unifies these two tasks remains elusive. Here we show that a single molecule can both store and decode information by using its own stochastic chemical dynamics. Confined within an individual protein nanopore, amino acid side-chains encode configurable memory tapes, with reactive cysteines representing '1' bits and unreactive residues representing '0' bits. Reversible thiol–selenosulfide exchange converts cysteine thiolates into inert selenosulfide adducts, producing nonlinear, history-dependent dynamics that enable data retrieval from seconds-long observation windows of the ongoing stochastic dynamics. All reactive memory tapes are identified with >97% bit-level and >90% overall 15-way classification accuracies in a 4-bit system, while producing tunable reservoir dynamics either by reactant concentration or tape composition. This work provides a minimal physical substrate that intrinsically unifies data storage with computation. Our findings may apply to any chemical reaction network with reversible transitions and a single-molecule readout, and may help elucidate design principles for stochastic chemical computing which are accessible only at single-molecule level.

KEYWORDS
molecular machines, chemical data storage, chemical computer

HFSP Reference Number: LTF-0040/2022-L
HFSP Award Category: Long-Term-Fellowship
HFSP Award Year: 2022

MENTAL 3D SPACE-TIME TRAVEL IN FISSION-FUSION ANIMAL SOCIETIES

Sara Torres Ortiz
University of Southern Denmark, Danmark
Cynthia Moss; Auguste Von Bayern; Herbert Peremans; Magnus Wahlberg
University of Southern Denmark, Danmark

ABSTRACT
Mental Time Travel (MTT)—the capacity to recall past experiences and anticipate future events—has traditionally been regarded as a hallmark of human cognition. The goal of this project was to investigate the cognitive mechanisms—Time Perception, Episodic Memory, and Episodic Foresight—that allow phylogenetically diverse species (dolphins, bats, and parrots) to solve survival challenges across space and time. We sought to determine how these animals manage information when immediate sensory input is no longer available, and how these biological principles can be translated into flexible biorobotic platforms. Using standardized delayed-response paradigms across phylogenetically diverse taxa—including dolphins, orcas, parrots, pinnipeds, bats, and otters—we identified a striking cross-species limitation: behavioral performance declined sharply when delays exceeded 18–20 seconds, suggesting a conserved boundary in working memory capacity. This constraint likely represents an evolutionary tipping point at which reliance on immediate sensory information becomes insufficient for adaptive decision-making. We then examined the mechanisms animals use to transcend this limitation. Dolphins demonstrated intentional encoding when anticipating future task demands, successfully transferring information into long-term memory for delays of up to 16 hours. Parrots recall incidentally encoded own actions at least after short delays but fail what-(when)-where episodic-like memory tests. Otters employed anticipatory problem-solving strategies that constrained future uncertainty before prey-tracking demands exceeded short-term processing capacity. Bats relied on spatial priors derived from long-term representations to navigate effectively when immediate sensory input was unavailable. To evaluate the translational relevance of these findings, we implemented biologically inspired memory architectures in biorobotic systems. Incorporating short-term buffers alongside weighted long-term priors enabled robotic agents to maintain performance under conditions of sensory decay, mirroring strategies observed in biological organisms. Our research suggests that vertebrates may face common short-term memory constraints of approximately 18–20 seconds. Across examined taxa, specialized cognitive mechanisms—prospective encoding in dolphins, anticipatory reasoning in otters, and reliance on spatial priors in bats—appear to function as a cognitive bridge, enabling adaptive behavior when immediate sensory traces rapidly decay. These strategies represent convergent ecological solutions to a shared processing challenge, allowing diverse species to solve complex time-dependent problems. By integrating these biological principles—particularly the dynamic weighting of stored representations against real-time sensory input—into biorobots and weakly supervised AI systems, we developed more robust autonomous platforms capable of operating under sensory uncertainty. Together, these findings advance a comparative framework for understanding how brains extend behavior beyond the limits of short-term retention and help identify a candidate temporal boundary between reactive responding and future-oriented control.

KEYWORDS
episodic memory, prospective memory, foresight, time perception, future-oriented cognition

HFSP Reference Number: RGP0045/2022

HFSP Award Category: Research Grant Program

HFSP Award Year: 2022

PIGMENT CONTENT, GENE EXPRESSION AND ORGANELLE FUNCTION IN VERTEBRATE PIGMENT CELLS

Athanasia Tzika
University of Geneva, Switzerland
Adrian Lutz[1]; Pierre-Yves Helleboid[2]; Pierre Beaudier[2]; Benjamin Palmer[3]; Devi Stuart-Fox[1]
[1]University of Melbourne; [2]University of Geneva; [3]Ben-Gurion University of the Negev

ABSTRACT
Animal colours arise from universal optical principles, yet their biological implementation is remarkably diverse. In ectothermic vertebrates such as reptiles, skin colour is produced by three main chromatophore types: melanophores containing light-absorbing melanins, xanthophores containing pteridins and carotenoids, and iridophores containing reflective guanine crystals. Interactions among these chromatophores generate the wide range of hues observed in reptile skin. While melanophores and iridophores have been intensively studied, the transcriptomic profile, biochemical composition, subcellular organisation, and physiological regulation of xanthophores remain comparatively understudied. Here, we present a comparative and mechanistic investigation of pigment composition and organisation in the skin of several agamid lizard species and multiple colour morphs of the corn snake, using the bearded dragon and corn snake as our focal models. Quantitative biochemical analyses demonstrate that pteridin concentrations are markedly elevated in the skin compared with other tissues, supporting a specialised and locally enhanced role in integumentary coloration. Cross-species and cross-morph comparisons reveal that variation in pteridin abundance correlates strongly with differences in colour saturation and hue. Beyond pteridins, we further characterise the contribution of riboflavin (vitamin B2) to reptile skin colour. Although not traditionally classified as a pigment, riboflavin is abundant in the integument and functions as a significant yellow chromophore. Our data support a central and previously underappreciated role for riboflavin in shaping colour output in combination with pteridins. To connect pigment composition with cellular architecture and molecular regulation, we integrate quantitative pigment analyses with transcriptomic profiling and high-resolution imaging. Transmission electron microscopy and expansion microscopy were performed independently to investigate xanthophore ultrastructure at complementary scales. By correlating organelle morphology with quantitative pigment content and gene expression patterns, we identify novel structural features of xanthosomes and previously unrecognised heterogeneity in their organisation. Finally, we investigate the physiological regulation of pigment production at the organelle level. Our findings indicate that intracellular redox state plays a previously underappreciated role in regulating pteridin accumulation and pigment organelle function in xanthophores. These results suggest that pigment production is not solely determined by biosynthetic capacity, but is tightly linked to the redox environment of the cell. Together, our results provide new insight into the biochemical and subcellular basis of reptile coloration and highlight xanthophores as dynamic, physiologically regulated contributors to vertebrate colour production.

KEYWORDS
animal coloration, reptiles, xanthophores, pigment biochemistry, physiological control of pigmentation

HFSP Reference Number: RGP0037/2022-302
HFSP Award Category: Research Grant
HFSP Award Year: 2022

SHINY SIGNALLING: THE PRODUCTION, DETECTION AND NEUROBIOLOGICAL PROCESSING OF BRILLIANT COLOURS

Casper VAN DER KOOI
University of Groningen, Neetherlands

ABSTRACT
Dynamic visual effects such as glossiness are taxonomically widespread and have evolved repeatedly across the tree of life. Examples can be found in brilliantly coloured butterflies, beetles, birds and flowers. The changeable nature of these organisms poses a challenge for reliable signaling, because for signals to be reliable, they should be consistent. Glossy visual effects defy that principle, because the bright, directional pulse of light that dominates their appearance is highly variable across space and time. We investigate the optical properties of dynamic visual effects and how these effects influence signal efficacy using bumblebees as a model of insect vision. Our optical studies reveal that dynamic visual effects can be caused by different optical mechanisms, ranging from flat cell structures in flowers to more complex multilayer structures in butterflies. Through behavioral experiments with artificial stimuli that mimic the spectral and spatial reflectance properties of glossy and matte floral surfaces, we demonstrate that glossiness enhances long-range detectability but compromises fine-scale color discrimination at close range. Glossiness thus poses an optical property by which organisms can attune their visual appearance independent of pigmentary properties, representing a functional trade-off between conspicuousness and signal reliability.

Video about our research: https://www.youtube.com/watch?v=FBQCf8wdWgI

KEYWORDS
optics, evolution, visual signalling, pollination, animals, plants

HFSP Reference Number: RGP0023/2023
HFSP Award Category: Research Grant Program
HFSP Award Year: 2023

MONSTERS OF THE APOCALYPSE: HOW FOSSIL TERATOLOGY HELPS IDENTIFY HYDROTHERMAL BRINES AS PLAUSIBLE TRIGGERS FOR ANCIENT MASS-EXTINCTION

Thijs Vandenbroucke
Ghent University (UGent), Belgium

Tim De Backer[1]; Iris Vancoppenolle[1]; Carolina Klock[1]; Patrick I. Mclaughlin[2]; Poul Emsbo[3]; Barry Lomax[4]; Cindy Looy[5]; Bas Van De Schootbrugge[6]
[1]Ghent University (UGent); [2]Illinois State Geological Survey; [3]United States Geological Survey; [4]University of Nottingham; [5]University of California Berkeley; [6]Utrecht Universiteit

ABSTRACT
The Silurian (c. 440 to 420 million years ago) is marked by several large extinction events that affected various clades of marine organisms, i.e., the Ireviken, Mulde and Lau events, named after the geological sites they were first described at, on the Swedish island of Gotland. These events, that helped shape the evolution of Earth's biosphere, were accompanied by large disruptions of the oceanic carbon cycle, recorded as positive stable carbon isotope excursions in contemporaneous carbonates. Despite this strongly held linkage, the fundamental cause-and-effect relationships that underpin these events have long remained obscure. Unveiling an important piece of the puzzle, our project has helped document a striking correlation between peak occurrences of severely malformed organic-walled fossil plankton and the onsets of the Silurian extinction events. Notably affected are chitinozoans, microfossil eggs of metazoans. Further analysis identified strong increases in redox-sensitive metals, toxic to marine organisms, in tandem with the records of teratology. By analogy with malformations of modern organisms triggered by toxic heavy metal contamination, this discovery confirms that chitinozoan deformations are fossilized in vivo responses to paleoenvironmental turmoil. Using these marked instances of metal-induced teratology, we identified pollution by metals as a contributing kill-mechanism to Silurian extinction, and linked the metal-spikes to redox changes that accompanied Ocean Anoxic Events. Furthermore, our integrated paleobiological-geochemical data, combined across these Silurian anoxic events, lend support to our nascent hypothesis that the massive venting of nutrient rich brines into the ocean during the formation of exhalative (sedex) Zn-Pb deposits, likely is their long-sought trigger.

KEYWORDS
Micropaleontology, teratology, OAE, Palaeozoic

HFSP Reference Number: RGP0066/2021
HFSP Award Category: Research Grant
HFSP Award Year: 2021

HOW A SINGLE CELL SHAPES A SHOOT

Teva Vernoux
ENS de Lyon, France

Laure Mancini[1]; Priyanka Patra[1]; Jonathan Legrand[1]; Guillaume Cerutti[1]; Yoan Coudert[1]; Odette Rozaleen[2]; Josepha Grewer[2]; Matias Zurbriggen[2]; Brendan Lane[3]; Robert Below[3]; Jordi Chan[3]; Richard Smith[3]; Lidor Shaar-Moshe[4]; Javier Jauregui-Lazo[4]; Muhammad Kamran[4]; Siobhan Brady[4]; Philippe Andrey[5]
[1]ENS de Lyon; [2]Heinrich Heine University Düsseldorf; [3]John Innes Centre; [4]UC Davis; [5]INRAE

ABSTRACT
Phyllotaxis, the regular arrangement of leaves around stems, is one of the most striking natural patterns and has evolved convergently in various plant lineages. In flowering plants, this pattern arises from a multicellular tissue at the shoot tip. In contrast, in the moss Physcomitrium patens, a single cell controls phyllotaxis. Successive rotations of the division plane of this single apical stem cell indeed directly determine moss phyllotaxis, with each apical cell derivative directly generating a leaf. This provides a unique system to investigate how the division of a single cell shapes the architecture of an entire shoot. We have established a live-imaging pipeline to extract quantitative cellular parameters at the moss shoot apex. Through this approach, we identified a geometric canalization mechanism by which the contribution of the geometry of the apical cell becomes increasingly important in controlling the position of the division plane during phyllotaxis establishment. This geometric control might also be sufficient to maintain phyllotaxis during subsequent stages. At the molecular level, the plant hormone auxin is a major regulator of phyllotaxis in flowering plants. Using micro-computed tomography, we have demonstrated that moss mutants affected in auxin efflux exhibit altered phyllotaxis, providing the first evidence that auxin was recruited independently for phyllotaxis control in mosses and flowering plants. We have found that auxin operates by regulating cell wall gene expression, composition, and likely mechanical properties. This has been further confirmed by performing single cell RNA sequencing of moss shoot apices and analyzing the developmental trajectories of the apical and daughter cells. We have also developed several optogenetic systems, specifically adapted to moss, enabling control of target gene expression with different light wavelengths. We have started to generate stable transgenic lines to misregulate hormonal activity and the expression of genes identified by single cell RNA-seq. Finally, we have generated a computational model of the shoot apex to probe if geometric and mechanical cues can aid in the positioning of the division plan of the apical cell. We have used it to analyze the contribution of geometry and differential growth in controlling the orientation of the apical cell division plane. Altogether, our work provides novel major insights into how the geometry of a single apical cell and its daughter cells, their physical properties and biochemical cues self-organize 4D patterns of division orientation and ultimately shape a shoot.

KEYWORDS
Developmental biology, plant, cell division orientation, predictive biology

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MOLECULAR CHECKPOINT FOR QUALITY CONTROL IN FLAGELLIN SIALYLATION

Patrick Viollier
University of Geneva, Switzerland

ABSTRACT
Sialic acid-based surface glycoconjugates govern cellular identity. Many bacteria sialylate flagellin with pseudaminic or legionaminic acid-derived glycans, often present in lipopolysaccharides and capsules (O-/K-antigens). How pseudaminic acid (Pse) synthesis and derivatization is coordinated is poorly understood. Here, we unveil the Caulobacter crescentus FlmEFX proteins as first Pse-derivatization and quality control checkpoint trageting the conserved UDP-GlcNAc (-N-acetyl glucosamine) dehydratase/epimerase FlmA which executes the first committed step of Pse synthesis. Heterologous reconstitution of FlmEFX-dependent flagellin sialylation in Escherichia coli and mutational analyses negative or positive checkpoint function of FlmX, a member of the unexplored WbqC family, and its role in Pse-derivatisation. Distinct surface mutations that uncouple FlmA from FlmX inhibition are in resides of allosteric Pse-binding pocket fomed when FlmA assembles into hexamers. We propose that FlmEFX implements a biosynthetic feedback to FlmA during abortive Pse derivatization, ensuring resourceful consumption of UDP-GlcNAc, a central precursor of envelope structures including LPS and peptidoglycan.

KEYWORDS
bacteria, flagellin, glycosylation, sialic acid, checkpoint, chemical derivatisation

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ILLUMINATING MESOPHYLL MICROSTRUCTURE IN THE CONTEXT OF LEAF MACROSTRUCTURE: LEAF GAS DIFFUSION DYNAMICS ACROSS DIVERSE FORMS

Nancy Walker
Yale University, USA

Aleca Borsuk[2]; Siul Ruiz[3]; Daniel Mckay Fletcher[4]; Anja Geitmann[5]; Craig Brodersen[1]
[1]Yale University; [2]The New York Botanical Garden; [3]University of Southampton; [4]Scottish Rural University College; [5]McGill University

ABSTRACT
Diffusion of CO2 through leaf intercellular airspaces is critical to its photosynthetic capacity by reaching all regions of leaf mesophyll, the photosynthetically active internal tissue of leaves. While mesophyll architecture is known to influence gas diffusion, diffusion dynamics within geometrically complex mesophyll structures remain poorly characterized. Most existing approaches rely on simplified one-dimensional approximations that do not capture the full three-dimensional diffusive physics. Here, we use homogenization, a mathematical upscaling technique, to quantify the three-dimensional diffusivity of leaf mesophyll from realistic geometries reconstructed via X-ray Micro-Computed Tomography across 73 plant species spanning distinct leaf forms (laminar, blade, and needle-like) and light environments (sun/shade).
Methodologically, we find that conventional scalar diffusivity metrics strongly correlate with overall diffusive capacity, but not preferential diffusive directionality (anisotropy), highlighting limitations in dimensionally reduced diffusivity estimates. Scientifically, our results show that diffusive magnitude and anisotropy are independently optimized according to leaf form, with variation linked to mesophyll organization patterns, and both diffusive traits exhibit plasticity with sun-shade adaptation. These results reveal fundamental links between leaf macro- and micro-structure, for which associated adaptive functional traits are likely strongly intertwined.

KEYWORDS
Plants, Biomechanics, Diffusion, X-ray Computed Tomography, Finite Element Modeling

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HFSP Award Category: Research Grant
HFSP Award Year: 2023

FUNCTIONAL ULTRASOUND IMAGING IN 4-MONTH-OLD INFANTS: A PILOT STUDY TOWARDS IMAGING EARLY LANGUAGE DEVELOPMENT

Kenta Watanabe
University of Tokyo, Japan

Sho Tsuji[1]; Leila Wehbe[2]; David Maresca[3]
[1]University of Tokyo; [2]Carnegie Mellon University; [3]Delft University of Technology

ABSTRACT
Understanding how the human brain supports language acquisition remains a central question in cognitive neuroscience. Established infant-friendly methods such as electroencephalography (EEG) and functional near-infrared spectroscopy (fNIRS) have limitations, including constraints in spatial or temporal resolution. Recent advances in ultrasound technology enables imaging of brain activity with a higher resolution and sensitivity than such methods. Functional ultrasound (fUS) imaging provides promising access to cerebral hemodynamics in human neonates. Neonatal fUS imaging takes advantage of the fontanelles – unfused gaps between cranial bones that permit ultrasound transmission to underlying cortical structures; however, its application beyond the neonatal period, when the fontanelles gradually start closing, remains largely unexplored. Here, we investigate the feasibility of fUS imaging in infants as old as four months of age, a developmental stage characterized by rapid growth in auditory and linguistic processing. We recorded fUS signals from ten 4-month-old infants while they watched a silent video. Measurements were obtained from multiple orientations and positions at the anterior and posterior fontanelles, which differ in both anatomical position and timing of closure, to assess their suitability for capturing cortical responses at this age. By extending fUS measurements beyond neonates, this study evaluates the potential of this technology to address unanswered questions in early language acquisition and will enable a wave of neuroscience studies into early language development.

KEYWORDS
functional ultrasound imaging, human infants, fontanelles, early language development

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HFSP Award Year: 2023

STRUCTURAL ONTOGENY OF PROTEIN-PROTEIN INTERACTIONS

Aerin Yang
EPFL (École Polytechnique Fédérale de Lausanne), Switzerland

ABSTRACT
Proteins carry out many of their biological functions through interactions with other proteins, yet how these interactions first arise and evolve remains a fundamental unsolved problem in biology. Some protein surfaces readily attract binding partners, including therapeutic antibodies and small molecule drugs, while other surfaces on the same proteins resist binding almost entirely. These "undruggable" surfaces represent a major barrier in medicine, yet the biophysical basis for why they differ from natural binding sites is not fully understood. To address this question, we used a synthetic proxy of protein-protein coevolution. We previously built this platform to remodel existing interfaces between small binding protein scaffolds, demonstrating that coevolution can diversify interface chemistry while preserving a common docking geometry (Yang et al., Science 2023)[1]. Here, we asked whether entirely new protein-protein interactions could emerge between surfaces with no natural history of binding to any ligand. Using a library-on-library selection strategy coupled to deep sequencing, we observed how new protein interfaces emerge from an unbiased starting point, capturing coevolutionary trajectories not readily accessible in naturally occurring proteins. We paired this platform with machine learning and epistasis analysis to estimate the fitness landscape associated with changes in binding affinity and specificity. In the system studied, our results reveal a notable difference between natural binding surfaces and surfaces that have not evolved to interact with other proteins. Natural binding surfaces converge on high-affinity complexes with a conserved docking geometry, consistent with a deep energy well favoring one binding solution. In contrast, coevolution on "silent" surfaces produces a spectrum of structurally distinct docking modes, each with lower affinity, reflecting a shallow energy landscape in which no single geometry is strongly favored. We identified six distinct structural families of synthetic protein complexes, together illustrating this plasticity directly (Yang et al., Science 2026)[2]. Epistasis analysis of the machine learning-estimated fitness landscape further identified specific "seed" residues predicted to initiate the first contacts between binding partners at the earliest stages of complex formation, proposed to act as nucleation points from which the full interface propagates through correlated, compensatory mutations. Together, these findings suggest that natural protein binding sites are evolutionarily pre-organized to support tight binding, while alternative surfaces are likely disfavored by evolution to avoid unwanted interactions. This framework provides evidence for a mechanistic hypothesis explaining why drugs and antibodies preferentially engage natural binding sites, with broad implications for drug discovery and protein engineering.
[1]https://www.science.org/doi/full/10.1126/science.adh1720
[2]https://www.science.org/doi/10.1126/science.adx6931

KEYWORDS
protein-protein interactions, protein engineering, synthetic coevolution, machine learning

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AN ANCIENT MONOAMINERGIC SIGNALING SYSTEM COORDINATES CONTRACTILITY IN A NERVELESS SPONGE

Rong xuan (Roy) Zang
Yale University, USA

Nawaphat Malaiwong[1]; Ling Wang[2]; Jamie Maziarz[1]; Kejue Jia[1]; Bernhard Drotleff[3]; Frank Stein[3]; Sean Liu[1]; Mah Noor[1]; C. Jackson Roberts[1]; Mandy Rettel[3]; Jennifer Schwarz[3]; Aissam Ikmi[3]; Shigeki Watanabe[4]; Robert Prevedel[3]; Noriko Funayama[5]; Michael O'Donnell[1]; Jacob Musser[1]
[1]Yale University; [2]College of Optical and Electronic Technology; [3]European Molecular Biology Laboratory (EMBL) Heidelberg; [4]Johns Hopkins Univerisity; [5]Kyoto Univeristy

ABSTRACT
Chemical neurotransmission was a key animal innovation, enabling multicellular coordination of physiology and locomotion. Sponges are early-diverging animals that lack neurons and muscles, yet still coordinate contraction and relaxation of their filter-feeding water canals. Here, we combine behavioral imaging, isotope tracing, heterologous expression, and functional proteomics to characterize the cellular players and molecular pathways involved in regulating whole-body movements for the freshwater sponge, Spongilla lacustris. Unlike canonical transmitters—including glutamate, acetylcholine, and catecholamines—used by other animals to regulate homeostasis, S. lacustris synthesizes tryptamine, phenethylamine, and tyramine as simple monoamines derived from amino acid decarboxylation to elicit distinct canal behaviors. We further demonstrate S. lacustris employs previously uncharacterized members of the pyridoxal-5′-phosphate-dependent enzyme and major facilitator superfamilies as decarboxylases and vesicular transporters, respectively, in secretory neuroid and metabolic cells for monoamine biosynthesis and transport de novo. Phosphoproteomics and label-free 3D imaging of S. lacustris reveal that tryptamine activates GPCR signaling and Rho GTPases, remodeling actomyosin and adhesion complexes in contractile canal epithelia to drive localized constrictions and whole-body deflations. Our findings support an evolutionary scenario in which molecular modules responsible for unicellular chemotactic crawling were co-opted into specialized cell types, thereby establishing the first monoaminergic circuits in animals to control tissue contractility, before undergoing further elaboration to support the neuromodulation of synaptic transmission in nervous systems.
https://www.biorxiv.org/content/10.64898/2026.02.14.704838v1

KEYWORDS
Evolution, behavior, neurotransmitter, actomyosin, coordination

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HFSP Award Year: 2023

ENZYME ENTRAPMENT AND VISUALIZATION IN METAL-ORGANIC FRAMEWORKS

Orysia Zaremba
Enamine Scientific Research Institute, Ukraine

ABSTRACT
Enzymes are a brilliant example of how nature has evolved highly efficient biological catalysts. Currently, synthetic analogues of enzymes are still far from achieving the catalytic efficiency and multifunctionality of the enzymes. Although enzymes are highly efficient catalysts, they are fragile, difficult to recycle and sensitive to adverse conditions, such as high temperatures, too high or too low pH values, toxic solvents, or even metal sources that could disrupt enzyme folding. In addition, the process of enzyme purification is resource-intensive, requiring significant energy, labour and time, thereby adding to the overall cost of production. To tackle enzyme fragility, a protection matrix is needed. Among various solid supports, metal-organic frameworks (MOFs) are excellent candidates because of their high crystallinity, structural rigidity, and extraordinary porosity, which allow the enzyme protection, the catalytic substrate transport across a MOF matrix, and the chemical tunability of the MOF/enzyme interface. It is also possible to synthesize MOFs under enzyme-compatible conditions and efficiently recycle them. Up to date, mainly purified enzymes have been combined with MOFs, while the crude enzyme extract, which contains enzymes along with cellular components, has not been explored as an enzyme source. The utilization of crude enzyme extracts could offer the advantage of retaining the structure and the activity of delicate enzymes that tend to deteriorate during the purification process. Another major limitation in enzyme entrapment research is the lack of methods for precisely imaging the distribution of enzymes and proteins entrapped in different solid materials. To address both the fragility of enzymes, their deactivation during purification, and incompatibility with MOF precursors during in situ entrapment, we directly use crude enzyme extracts obtained from cell lysis instead of pure enzymes by entrapping them in MOFs structures, as well as the development of enzyme-compatible MOF syntheses that take into account compatibility of metal salts with enzymes. We focus on three enzyme types with varying sensitivities: aldoxime dehydratase, imine reductase and lipase. We evaluate the effects of different metal sources (Al, Fe, Co, Ni, Cu, Zn), their oxidation state and counterions, as well as MOF synthesis parameters on the enzyme stability and activity during their entrapment in the MOF structures. Based on these findings, we optimize protocols for enzyme entrapment in Fe-MIL-88A, Fe-MIL-100, Zn-MOF-74, Zn-ZIF-8 and develop a fast-aqueous room temperature synthesis of Al-MIL-53. Furthermore, a new methodology of visualizing enzymes in a MOF matrix using a probe corrected FEI Titan microscope has been developed to improve our understanding of enzyme distribution within a MOF scaffold. Investigation of the biocatalytic performance of the enzyme@MOF biocomposites suggests that enzyme entrapment in MOFs using crude enzyme extracts can effectively maintain enzyme activity and stability in various catalytic reactions.

KEYWORDS
enzymes, MOFs, entrapment, visualization, biocomposites, biocatalysis

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HFSP Award Category: Research Grant
HFSP Award Year: 2024

MICRO/NANOSCALE THERMAL CONTROL OF A SINGLE LIVING CELL ENABLED BY ALL-OPTICAL DIAMOND HEATER-THERMOMETER

Vadim Zeeb
NanThermix SA, Switzerland

ABSTRACT
Despite the constantly growing interest in local thermodynamic phenomena in living cells, intracellular thermodynamics is still a matter of guesses and controversies due to the clear absence of reliable intracellular temperature control methods functional at micro and nanoscale. The long-lasting paradigm in Life Science usually neglected theoretical assumptions of possible ultra-local micro/nanoscale temperature gradients existence bound to heat dissipation related to intracellular thermodynamics. For example, electrical currents through open ionic channels operating under membrane voltage around 70mV/ 7nm corresponding to electrical field strength of 10 000 000 V/m - usual in CERN particle accelerator experiments. Such currents by definition must produce heat since intracellular media is not an electrical superconductor. Calcium binding to high affinity buffers as well as ATP- dependent processes including ion pumping, inevitably must produce heat as well. Surprisingly, by peculiar nature's selection - the water constituting the base of intracellular media is the champion for two main thermodynamic parameters: it has the highest heat capacity and the lowest heat conductivity. In reality the heat conductivity coefficient of intracellular media is 5 to 7 times smaller than that of the water which is in fact the value of good natural thermal insulator asbestos. Basing on Fourier's Law of heat transfer by conduction the low heat conductivity is in favor for the expectation that even tiny, intracellular heat sources of realistic heat power can produce steep steady-state or transient temperature gradients up to tens of degrees Celsius at nanoscale nearby active ion pumps, open ion channels, etc. This needs the updated view for macroscopic parameter - temperature for nanoscale volumes in aqueous media relying on the notion of local equilibria while taking into account that macroscopic limitation implies macroscopic amounts of molecules, but not necessarily macroscopic size of the system. We present a new methodology – micro/nano scale control/clamp of a temperature gradient profile in aqueous media by a fully optical Diamond Heater-Thermometer (DHT) which has striking functional similarities with Patch Clamp operating with voltage and current whereas DHT controls temperature and heat flow at a predefined point at micro/nanoscale (sub degrees Celsius and sub milliseconds resolution). The fundamental importance of the new way of ultra-local temperature control consists in the fact that it practically introduces this parameter into cellular physiology, namely into the privileged set of ultra-local parameters of the electrochemical potential equation similarly to the transmembrane electric potential and ionic concentrations. This all-optical new tool is compatible with Patch Clamp and Ca2+ imaging and unambiguously tames the temperature at the nanoscale, the last parameter of the electrochemical potential which still escaped its precise ultra-local experimental control in intracellular compartments.

https://pmc.ncbi.nlm.nih.gov/articles/PMC12821019/
https://www.nature.com/articles/s41598-023-35141-4
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KEYWORDS
nanoscale, temperature, control, cell

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The International Human Frontier
Science Program Organization
12 Quai Saint Jean
67000 Strasbourg France
www.hfsp.org