POSTERS SESSIONS with Abstracts

Heidelberg, Germany

October 16-18 2026

Center for Organismal Studies
University of Heidelberg

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Posters Session 1

Wednesday 16th, 2026

19.10 - 21.00

16.1 Jun Ru Lee - European Molecular Biology Lab, Heibelberg

Title: The interplay between developmental timing and tissue morphogenesis in teleost early embryogenesis

Early embryogenesis in metazoans follows a conserved sequence of events: fertilization, rapid and synchronous cleavages, slowing and desynchronization of cell cycles, exit from pluripotency, and eventually gastrulation marked by cell fate specification and coordinated tissue movements. These processes require the coordination of cellular and molecular program, such as the mid-blastula transition (MBT) and zygotic genome activation (ZGA), which guide the cell cycle elongation, desynchronization, and the onset of cell motility. Although these developmental programs are broadly conserved, they frequently result in diverse modes of morphogenesis across species. In this work, we hypothesize that heterochrony in the MBT and ZGA contribute to tissue morphological diversity at the onset of vertebrate gastrulation. To address this hypothesis, we employ teleost fish as a comparative model system. While teleost embryos share similar embryo structures, their morphogenetic processes, including epiboly and gastrulation, varies across species. We mapped cell cleavage dynamics across three species: zebrafish, killifish and medaka. We found that cell cycle elongation follows a hyperbolic growth, driven by resource consumption and Michaelis-Menten kinetics, which describe the cell duplication rates as a function of resource concentration. In this growth pattern, a mathematical singularity coincides with the cell cycle generation when gastrulation begins. Notably, cell cycle length in killifish and medaka reaches the singularity one cycle earlier than in zebrafish, suggesting a heterochronic shift in gastrulation timing between species. We further confirmed this heterochronic shift in gastrulation through the analysis of organizer gene expression and dorsal organizer compaction.

We propose that the heterochronic shifts of ZGA and MBT influence the spatial and temporal patterns of tissue mechanics, leading to distinct morphogenetic outcomes. Taken together, our results suggest that changes in the timing of early development alter the tissue mechanics, which in turn drives the diversity in teleost morphogenesis.

16.2 Carole Baumann - HIPS Saarbrücken

Title: An Integrated Zebrafish Platform for Anti-Infective Discovery, Toxicity Assessment, and Translational Research

Background

Zebrafish (ZF) larvae are a powerful vertebrate model for infection, pharmacology, and toxicology studies. Their optical transparency, conserved innate immune system, and suitability for high-throughput in vivo analysis enable real-time investigation of host–pathogen interactions and compound effects. Furthermore, their use prior to 5 days post-fertilization supports the 3R principles (Replacement, Reduction, Refinement) in preclinical research.

Methods

At the Helmholtz Institute for Pharmaceutical Research Saarland (HIPS), we have established an integrated zebrafish platform for the in vivo characterization of bioactive compounds and infectious disease processes. Transgenic reporter lines, combined with fluorescence microscopy, enable real-time visualization of host–pathogen interactions and organ-specific toxicities. Infection models employing clinically relevant pathogens facilitate the evaluation of antimicrobial efficacy, disease progression, and pharmacodynamic responses.

Results

The HIPS zebrafish platform supports anti-infective drug discovery by integrating efficacy, mechanistic, and safety studies within a single vertebrate model system. We present infection models for clinically relevant pathogens, including Acinetobacter baumannii and Staphylococcus aureus, which have been applied to evaluate natural products and other therapeutic candidates in vivo. The platform further enables the investigation of virulence determinants and host defense mechanisms.

To address the major causes of drug attrition, the platform incorporates early-stage toxicity profiling through predictive zebrafish models to assess cardiotoxicity, hepatotoxicity, nephrotoxicity, ototoxicity, and developmental toxicity. These assays are complemented by advanced bioanalytical methods that provide insights into compound uptake, distribution, metabolism, and host responses.

Conclusion

This integrated zebrafish platform provides a versatile framework for anti-infective research, combining efficacy testing, mechanistic studies, pharmacological characterization, and toxicity assessment in a 3R-compliant vertebrate model. By enabling the simultaneous evaluation of therapeutic efficacy, exposure, and safety, it supports the preclinical development of next-generation therapeutics.

16.3 Caroline Zandecki - KU Leuven

Title: The African turquoise killifish as a preclinical model for drug discovery in aging and regeneration

Age remains the strongest risk factor for the onset and progression of neurodegenerative diseases such as Alzheimer's and Parkinson's diseases, as well as the main factor leading to worse outcomes in conditions like traumatic brain injury and sarcopenia. To efficiently include this variable in the search for new druggable targets and therapies, we employ the African Turquoise killifish (Nothobranchius furzeri), an established vertebrate model in gerontology research.

The killifish offers unique advantages in aging research due to its exceptionally short lifespan (4-6 months in laboratory conditions) and its ability to recapitulate many hallmarks of mammalian aging at phenotypic, cellular and molecular levels.

Contrary to other regenerative-competent species, the killifish exhibits clear signs of spontaneous neurodegeneration and an age-dependent decline in regeneration capacity. These features make the killifish a favourable model to provide valuable insights into the underlying mechanisms of aging and the development and treatment of age-related diseases.

Building on these characteristics, we are developing a robust preclinical testing platform to assess the efficacy of both repurposed and novel compounds aimed at promoting healthy aging and tissue regeneration. Using the shortest-lived N. furzeri strain GRZ-AD, the platform integrates key readouts such as survival analysis, behavioural assessments, body mass index, spinal curvature evaluation, and muscle atrophy measurements. To assess compound efficacy in a regenerative context, or to include the factor neurodegeneration, we apply optimized injury models, including traumatic brain injury. At the molecular and cellular levels, we examine markers of cellular senescence, inflammation, neurodegeneration and muscle denervation, using a toolkit of readouts ranging from qPCR and spatial techniques to in-depth single-cell omics approaches.

Taken together, by leveraging the unique features of the African turquoise killifish, our platform offers a scalable model system to explore mechanisms of aging and regeneration, aimed at evaluating promising compounds and advancing therapeutic strategies in the field.

16.4 Sabrina Pfitsch - IBCS-BIP Karlsruhe

Title: Deciphering the Molecular Logic of Neurogenesis in the Zebrafish Spinal Cord – With Focus on the V2-domain

The nervous system is intricately organized by billions of neurons that orchestrate complex physiological responses. However, the detailed molecular mechanisms governing neuronal diversity remain incompletely understood. We focus on the zebrafish spinal cord, a model system that closely mirrors neuronal development in the mammalian spinal cord. Along the dorsal-ventral axis, antagonistic morphogenetic signals establish 11 distinct precursor domains, including the p2 domain, which gives rise to V2a, V2b, and V2s interneurons.
However, the mechanisms driving V2b and V2s subtype differentiation remains unclear.
To elucidate the molecular determinants of p2 progenitor cell diversification, we utilized the zebrafish vsx1:GFP transgenic line, which labels subpopulations within the p2 domain from an early developmental time point, enabling their characterization by single-cell RNA sequencing. Our analysis revealed insm2 as a potential regulator of precursor V2b differentiation, while foxp2 emerged as a specific late marker of V2s cells. To decipher the function of these markers in V2 development and subtype specification, we are currently performing loss-of-function assays using CRISPR/Cas9. By decoding the molecular logic of p2 progenitor diversification, our research advances the understanding of spinal cord development and the genetic regulation of neuronal diversity.

16.5 Aristides Arrenberg - University of Tübingen

Title: Electrical stunning as a humane euthanasia alternative

Refinement of euthanasia methods is essential for improving zebrafish welfare in research. Current methods, such as tricaine overdose and hypothermic shock, have key limitations, including aversive effects, variable efficacy, and prolonged time to death.

Our recent work in larval zebrafish demonstrated that electrical stunning provides a rapid and reliable alternative: under optimized alternating current conditions, larvae lose equilibrium in under 1 second, show immediate loss of sensory-evoked brain activity, and achieve 100% mortality with minimal morphological damage compared to other electrical parameter sets. These findings support electrical stunning as a promising refinement strategy for zebrafish euthanasia. In addition to the published work, we currently design electrical stunning chambers for adult fish and we investigate the downstream effects of electrical stunning on tissue integrity and physiological stress responses.

This integrated behavioral, neurophysiological, histological, and biochemical framework aims to determine not only whether electrical stunning is effective and practicable but also whether it offers a more humane euthanasia method.

Our work positions electrical stunning as a practical and scalable alternative to current zebrafish euthanasia procedures. Our results provide hitherto lacking evidence as well as suitable equipment and protocol design, and thereby advance refinement of zebrafish experimentation and husbandry under the 3Rs principles.

16.6 Rama Mausam - Justus Liebig University

Title: IL-11 signaling couples transcriptional and epigenetic stromal cell reprogramming to enable cardiac regeneration and suppresses scar formation.

Unlike the adult human heart, which forms non-functional scar tissue after injury, the adult zebrafish heart can regenerate functional cardiac muscle with minimal scarring. Effective cardiac regeneration depends on the orchestrated and precise integration of various cell types, where their interactions prompt the temporary activation of regenerative cell states. To gain a deeper understanding of the specific extracellular signals and intrinsic epigenetic frameworks that trigger these transient functional cell states, we characterized the chromatin landscapes of cardiac stromal cell populations within the regenerating heart from wildtype and mammalian like scar inducing, non-regenerative interleukin-11receptor (il11ra) deficient hearts at single-cell resolution. Our data reveals cis-regulatory elements and trans-regulatory factors that drive cell-specific gene expression patterns. Notably, we identified cell type specific Interleukin-11 dependent and independent chromatin remodelling of regulatory elements as early as 24hrs post injury and 96 hrs post injury. Our analysis reveals regeneration specific interleukin-11 dependent remodelling of elements in proximity to il11a, aldh1a2, lepb which are known regulators of cardiac regeneration in zebrafish.
Moreover, we identified elements with higher accessibility in fibrotic il11ra mutant hearts linked to genes acvr1l and sox9b which are responsible for fibrosis in mammals. In addition, motif enrichment analysis revealed enrichment for Stat transcription factors binding sites which act downstream of il11ra in regenerating stromal cells when compared to our fibrotic model which shows an enrichment of egr1, a known key regulator of fibrosis in mammals. Our findings thus underscore the critical early role played by endocardial cells (EndoCs) and epicardial-derived cells (EPDCs) in the cardiac regeneration process in zebrafish, and provides key insights into how these animals escape a fibrotic response.

16.7 Mathilde Hoareau - KIT Karlsruhe

Title: Deciphering the molecular mechanisms underlying successful brain regeneration in zebrafish

The brains of humans and other adult mammals have a limited capacity to produce new neurons and repair damaged nerve tissue. Adult zebrafish, on the other hand, form new neurons in abundance and repair brain damage very efficiently. These properties have made the zebrafish a powerful model for studying the cellular and molecular mechanisms underlying neurogenesis and regeneration in adult vertebrates. Recent studies have revealed that subsets of larval zebrafish neural stem cells (NSCs) already display transcriptional signatures similar to adult quiescent NSCs, indicating that features of the adult neurogenic program may emerge early during development. This suggests that larval zebrafish could be a highly effective and accessible model for investigating brain regeneration. However, it remains unclear whether regenerative responses in larvae recapitulate those observed in adults and whether they rely on conserved molecular pathways and regulatory mechanisms. To address these questions, we developed a telencephalic injury model in 4 days post fertilization zebrafish larvae. Using this model, injured and uninjured telencephalic tissues were collected at multiple post-injury time points to generate bulk RNA-sequencing libraries. This dataset will provide a comprehensive characterization of the larval zebrafish regenerative response and will enable direct comparison with existing adult datasets. Future work will integrate single-cell RNA-seq (gene expression profiling), ATAC-seq (chromatin accessibility analysis) and CAGE-seq (transcription start site identification) approaches to identify injury-responsive genes and their regulatory elements. To elucidate the mechanisms underlying the expression of these transcripts, we will select and characterise their potential regulatory sequences by transgenesis, sequence deletion or mutation of their potential regulatory domain crucial for their expression in neural stem cells of the telencephalic ventricular zone using the CRISPR-Cas9 methodology. A better understanding of the regenerative mechanisms occurring in the zebrafish brain may provide a foundation for developing new therapies to combat the debilitating consequences of brain injury, stroke, and neurodegeneration.

16.8 Eric Bienert - Justus Liebig University

Title: Restraining Inflammation to enable Regeneration: Junb as a key Il11-Stat3 effector in the zebrafish (Danio rerio) tissue regeneration model

With few exceptions, the predominant mode of tissue repair in mammals is scarring due to limited regenerative capabilities. Consequently, injuries result in reduced organ function and are a major contributor to severity and morbidity in human diseases. While low regenerative capacity is widespread among mammals, it is not a general trait of vertebrates. The zebrafish (Danio rerio) is known for high regenerative capacity, where damage to organs or extremities leads to complete regeneration and refunctionalization of the damaged tissue.

Interleukin-11 (Il-11) signaling with the downstream target Junb was identified as a key regulatory pathway in zebrafish regeneration. Junb functions as part of the activator protein 1 complex (AP-1) by dimerizing with proteins from the Fos, ATF or Maf family to bind DNA and regulate transcription. Here we show that injury in the zebrafish leads to increased expression of junba and junbb. This increase is absent in non-regenerative fish lacking the Interleukin 11 receptor (il11ra-/-) revealing Junba/b as targets of the canonical Il-11-stat3 signalling axis.Using junba and junbb mutant alleles, we investigate the role of Junb utilizing the zebrafish larval fin fold regeneration model. Employing genetic models and pharmacological interventions, we prove its importance for regenerative processes in zebrafish through regulation of post injury inflammation.

In junb-/- larvae, additional to impaired regeneneration, tissue damage also leads to severly increased inflammatory markers when compared to regenerative wildtypes.

Intriguingly high inflammation is also visible in mammals in the mouse myocardial infarction model, suggesting a connection between high inflammation and the absent ability to regenerate tissue.

Notably, pharmacological intervention with anti-inflammatory drugs restored regeneration in junb-/- confirming Hyperinflammation as the cause for impaired regeneration in Junb deficient zebrafish larvae.

16.9 Antonia Hauth - COS, University of Heidelberg

Title: Simultaneous chromatin accessibility and transcriptomic profiling of Neural Crest Cells in zebrafish and medaka using SHARE-seq

Neural crest cells (NCCs) show a remarkable potential for differentiation that was likely central to the evolution of the vertebrate "new head" and the diversification of over 66,000 described vertebrate species occupying nearly every ecological niche. During NCC differentiation and migration, large-scale changes in gene expression and chromatin architecture must occur to allow for the establishment of the diverse progeny. While single-cell chromatin and transcriptomic analyses of NCCs have provided important insights into lineage-specific enhancer dynamics, existing studies rely on computationally integrated datasets from separate assays or sorted NCC populations, leaving the direct, cell-by-cell coupling between chromatin state and gene expression during NCC specification unresolved in a whole-organism context. To address this, we are optimizing the combinatorial-indexing-based SHARE-seq multimodal single-cell workflow for use in teleosts. SHARE-seq allows simultaneous readout of the transcriptome (scRNA-seq) and open chromatin landscape (scATAC-seq) from the same cell, enabling direct inference of the cis-regulatory elements driving gene expression decisions at the moment of cell fate commitment. By testing a combination of methodological changes, we aim to identify the optimal approach for preserving both RNA and chromatin integrity in embryonic teleost tissue. Applying SHARE-seq to zebrafish and medaka embryos will generate the first comparative whole-organism, single-cell multimodal chromatin accessibility and gene expression atlas of vertebrate development, capturing the regulatory dynamics of NCC emergence and differentiation across all their progeny simultaneously.

16.10 Bettina Welz - EMBL Heidelberg

Title: Genetic variation shapes developmental diversity across medaka strains at single-cell resolution

Phenotypic diversity spans multiple levels of biological organization and arises through the interplay of genetic and environmental factors. Embryonic development represents a critical window during which variation must be balanced with robustness. Developmental programs must reliably generate functional organisms while retaining plasticity to accommodate environmental change. Understanding how developmental programs respond to environmental variation is particularly important in externally developing teleosts, where factors such as temperature strongly influence developmental rate and cellular processes. Yet, how naturally occurring genetic variation shapes diverse developmental trajectories, and how these trajectories correlate with environmental sensitivity, remains poorly understood.

Medaka (Oryzias latipes) is a powerful model for studying phenotypic diversity, offering hundreds of wild-derived inbred strains from distinct latitudes, capturing naturally occurring genetic variation associated with climatic adaptation. Here, we examine how developmental progression differs among five wild-derived medaka inbred strains. Using single-cell RNA sequencing with combinatorial indexing and embryo-specific hashing, we quantify cell-type composition and transcriptional dynamics across embryogenesis in more than 100 embryos. Our analyses reveal strain-specific differences in developmental rate and transcriptomic ages across cell types, indicating that genetically distinct strains follow divergent developmental dynamics even under common environmental conditions, suggesting that natural genetic variation shapes the tempo and coordination of embryonic programs.

To investigate how temperature shapes developmental trajectories, we expose the reference strain Cab to a wide temperature gradient (16–34 °C) for six days. This resulted in embryos spanning a broad range of developmental stages, reflecting temperature's role as a modifier of developmental speed.

Comparing these to a standard-temperature reference dataset allowed us to disentangle temperature-induced transcriptional changes from stage-dependent gene expression, identifying genes associated with thermal responses. Together, this establishes a baseline which we will expand to investigate how strain-specific trajectories interact with environmental variation, aiming to identify mechanisms that confer developmental robustness and plasticity under changing climates.

16.11 Sophia Papadopoulou - Max Planck Institute for Heart and Lung research

Title: Investigating the role of fibroblast genes at the onset of cardiac fibrosis in zebrafish

Cardiac fibrosis (CF) is a common pathological process underlying many cardiovascular diseases, including atrial fibrillation and hypertension. CF is characterized by excessive deposition of extracellular matrix components such as collagens, predominantly produced by activated fibroblasts. Although the clinical importance of CF and the role of fibroblasts are established, the molecular mechanisms of fibroblast-specific genes driving the onset and progression of CF remain unclear. To dissect these mechanisms, we are using our recently established gain-of-function (GOF) zebrafish model that constitutively overexpresses the sodium channel gene scn5lab in atrial cardiomyocytes. These transgenic zebrafish exhibit increased collagen deposition from 1 to 3 months post-fertilization (mpf) accompanied by impaired cardiac morphology and function. To elucidate the underlying molecular mechanisms and identify fibroblast-specific subpopulations involved in the onset of CF (i.e., 1 mpf), we performed single cell RNA-sequencing (scRNA-seq) to compare atrial tissue from control and fibrotic hearts. Single cell RNA sequencing revealed multiple transcriptionally distinct fibroblast populations whose marker genes were also upregulated in the atria of fibrotic hearts. A literature review indicated that these genes are implicated in extracellular matrix-stabilization and homeostasis.

Based on these findings, we hypothesize that these genes contribute to the onset of CF. To test this hypothesis, we plan to generate molecular tools, including mutant and knock-in lines to investigate their role in fibroblast activation and CF progression in vivo.

16.12 Amrutha Manikandan - Justus Liebig University

Title: Divergent IL-11 and IL-6 signalling shape cardiac injury outcomes in regenerative vs. non-regenerative vertebrates

In most mammals, cardiac injury typically results in permanent fibrotic scarring. In
contrast, zebrafish exhibit complete, scar-free regeneration of damaged cardiac tissue. Interleukin 11 (IL-11) has previously been identified as a critical mediator of the regenerative response in zebrafish, whereas in mammals it is commonly associated with pro-fibrotic signalling. However, the dynamics of IL-11 expression in early and late phases of the injury response in mammals remains poorly understood. To investigate this, we performed left anterior descending (LAD) artery ligation in wild-type C57BL/6 mice and collected heart tissue for bulk RNA sequencing at 6, 24, and 96 hours postinjury. In parallel, wild-type zebrafish hearts were subjected to cryoinjury, followed by RNA sequencing at 24 and 96 hours. Interestingly, IL-11 was upregulated in both mice and zebrafish at 24 hours post-injury. However, the IL-11 receptor (Il11ra1) was significantly downregulated only in mice. Notably, IL-6 expression and signalling was strongly upregulated at all time points in mice, but not in zebrafish. By 96 hours, fibrotic pathways were activated in mice but were not upregulated in zebrafish. Overall, our data
show that although IL-11 is upregulated in both species following cardiac injury, the downstream signalling and outcomes are fibrosis and regeneration in mice and zebrafish respectively. Furthermore, proteomics analysis revealed that IL-11 alone did not significantly upregulate fibrotic protein expression after 24 hours of treatment in mouse epicardial fibroblasts.
However, Il11ra1-/- cells displayed reduced expression of fibrotic proteins and downstream signalling. This highlights a dichotomy in IL11ra1-mediated signalling in regenerative versus non-regenerative vertebrates. In particular, IL6 as well as altering IL11Ra1 signalling in the mouse injury response, represent promising therapeutic targets to promote regenerative repair in the mammalian heart.

16.13 Chiau Yuan Hsu - COS, University of Heidelberg

Title: Medaka Embryonic Stem Cells Spontaneously Acquire Retinal or Forebrain Fate Without Directed Differentiation

The developmental fate of a single pluripotent stem cell in the absence of exogenous signaling cues remains a fundamental question in developmental biology. In this study, I cultured medaka pluripotent cells at low density in minimal media and generated retinal organoids from medaka embryonic stem cells. To maintain cells in suspension and prevent aggregation or substrate attachment, I used hydrogel immobilization and shaking-flask culture conditions. I monitored the adoption of retinal fate by Rx3::GFP (early retinal progenitors) and Rx2::RFP (retinal stem and progenitor cells) transgenic reporters, allowing to address gene expression dynamics in individual stem cells and retinal organoids over time. I found that a subset of suspended embryonic stem cells, as well as a subset of retinal organoids, autonomously initiated Rx3 expression, indicating specification toward a forebrain or retinal lineage. In contrast, Rx2 expression was observed only in retinal organoids and was absent in suspended stem cells, arguing that Rx2 activation requires cell-cell interactions or additional extrinsic signals. These findings indicate that medaka embryonic stem cells autonomously initiate early retinal or forebrain specification under minimal conditions, whereas progression toward further retinal differentiation requires external cues.

16.14 Mirjam Layer - COS, University of Heidelberg

Title: Simultaneous dual-fate mapping to resolve convergent differentiation in vivo

Throughout embryogenesis, multiple tissues are formed via the complex interplay of distinct embryonic lineages. And in some cases, multiple lineages converge to produce the same cell type. For example, the vertebrate skull is comprised of endochondral bone from neural crest (NC) and mesodermal origins. The majority of evidence for dual-lineage contributions to the same cell population is based on inferences from fate mapping one lineage at a time. Moreover, previous studies in our lab have revealed that the gene expression programs of cranial chondrocytes reflect their embryonic origin, suggesting lineage-specific mechanisms of fate specification. To systematically characterize the dynamics of dual-lineage convergence in vivo, I am generating a simultaneous, dual-recombinase system in zebrafish. First, we are driving recombinase enzymes (Cre and flipase) under endogenous control of lineage-specific transcription factors using CRISPR-Cas9 knock-in approaches. In combination with the zebrafish pIGLET safe-harbor system, we will label NC and mesoderm lineages with unique fluorophores downstream of a recombination cassette. We aim to resolve the cellular and molecular dynamics of cell fate convergence, both in our main tissue of interest, the embryonic skull, and embryo wide by combining live imaging and single cell transcriptomic readouts. We anticipate that in addition to resolving key questions about when and how fate convergence is achieved in the embryo, these tools will be broadly useful to the zebrafish community.

16.15 David Czimer - ELTE Eötvös Loränd University, Hungary

Title: A breath of fresh air: evolution and development of breathing strategies in the paradise fish

Demand for oxygen has been a main driver for evolution in Metazoans. While complex multicellular life first emerged in aquatic environments, the rise of atmospheric oxygen facilitated the repeated evolution of air-breathing strategies, granting access to this abundant and relatively stable resource. Indeed, recent research suggests that air-breathing evolved in fishes over 80 times independently. Paradise fish (Macropodus opercularis) is an air-breathing freshwater fish species, with a signature labyrinth organ (LO) capable of extracting oxygen from the air that helps adult fish to survive in hypoxic environments. The appearance of this evolutionary innovation in the group of anabantoid fishes resulted in anatomical innovations, and also contributed to the emergence of species-specific behaviors, such as parental care. While at the beginning of the 20th century zoologists were intrigued by the structure and function of the labyrinth apparatus, ultimately this research program petered out due to the lack of adequate molecular tools to address the questions related to the development of the LO. Using some of the latest technological developments, we revisit these questions to reveal the molecular and cellular landscape of a true evolutionary innovation. We aim to use a comparative stratgey, not only to understand how paradise fish evolved the LO, but also to reveal if this organ shows convergent features with orthologous air-breathing structures in other species.

16.16 Anastasja Kovacevic - Leibniz Institut für Altensforschung

Title: Role of rRNA genes in N. furzeri aging

In invertebrate systems, the link between aging and RNA Polymerase I (Pol I) has been elucidated. Activity of Pol I results in pre-rRNA production and takes place in the nucleolus whose size reflects the rate of ribosome production, and consequently, the cell's growth rate and energy demands. In invertebrate systems, suppression of Pol I activity, reflected by reduced pre-rRNA level, has been associated with nucleolar size shrinkage and lifespan extension, mediated by the inactivation of rrn3 (tif-1A), a key rate-limiting Pol I transcription factor. To investigate if this link exists in vertebrate systems, Nothobranchius furzeri (turquoise killifish), the shortest-lived vertebrate model maintained in captivity, was employed as a suitable model. Characterization of the MZCS-08/122-rrn3- line, showed that there is a reduction of both rrn3 and pre-rRNA levels in heterozygous mutants while homozygous mutants are embryonically lethal. Nevertheless, rrn3+/- female hepatocytes did not exhibit decrease in nucleolar size upon Pol I suppression. Moreover, a trend towards lifespan extension in male heterozygous mutants has been observed. Since we think that mutation in heterozygous state is not enough to cause drastic changes in the lifespan, next step is crossing MZCS-08/122-rrn3- line with the line with altered polr1a level (gene encoding for the largest subunit of Pol I) and that will allow further reduction of pre-rRNA level. On the other hand, a detrimental effect of promoted Pol I activity on lifespan, upon rrn3 overexpression, was observed in C. elegans. To investigate this further in N. furzeri, a line with overexpressed rrn3 was successfully created and it will be used as a model for further investigation of how rrn3 and pre-rRNA upregulation influence organismal lifespan and health span. Furthermore, nucleolar biology investigation in turquoise killifish has shown a sex-specific difference of nucleolar size in liver tissue with females having larger nucleoli than males.

16.17 Rima Siauciunaite - KIT Karlsruhe

Title:Circadian Control of Cholesterol Metabolism in Glioblastoma

In a healthy brain, circadian clocks ensure that metabolism follows a daily rhythm. In cancer, however, this timing is often disrupted - but it is still unclear whether this loss of timing contributes to tumour development. This study investigates whether glioblastoma (GBM) loses circadian control over the mevalonate pathway, which is a key pathway for cholesterol synthesis that supports both normal glial function and tumour growth.

We used a combination of an in vivo zebrafish brain model, which preserves natural circadian rhythms, and human GBM models grown in 2D and 3D cultures. After synchronising the circadian timing, we measured the temporal expression of key mevalonate pathway genes (HMGCS, HMGCRA, FDPS) and assessed tumour-associated behaviours such as proliferation, invasion, and stem-like cell features. We also investigated the role of the clock by pharmacologically targeting core circadian regulators (CRY and REV-ERB).

We found that cholesterol biosynthesis genes exhibit distinct daily rhythms in non-malignant zebrafish brain tissue. In contrast, GBM models lose this temporal organisation. Disrupting circadian regulators further weakens this control and enhances tumour-like behaviour.

These findings suggest that glioblastoma is characterised not only by metabolic changes, but also by a loss of metabolic timing. Restoring circadian control could therefore represent a new approach for targeting tumour metabolism.

16.18 Eliana Stanganello - Tron Gmbh Mainz

Title:Imaging Cellular Dynamics in Zebrafish: From Cytotoxicity to Therapeutic Uptake

Immunotherapy has significantly improved survival in several cancer types that previously responded poorly to standard treatments. However, many malignancies still show limited benefit from current immunotherapeutic approaches, underscoring the need to develop new strategies and optimize existing ones.
In our lab, we integrate high-content imaging with the zebrafish model system to visualize biological processes across spatiotemporal scales. This combination enables us to study cellular interactions, dynamic behaviors, and intracellular trafficking of delivered cargo with high spatial resolution and temporal precision, both in vitro and in vivo. By coupling these imaging approaches with dedicated analytical pipelines, we generate quantitative datasets that allow us to track condition-specific differences and evaluate test items over time.
Our research focuses on cytotoxicity assays that capture the dynamic interactions between effector and target cells. Using these platforms, we investigate the performance of NKs, antigen-specific T cells, CAR T cells, and bispecific antibodies. In parallel, we monitor internalization processes to visualize and understand the uptake of lipid nanoparticles (LNPs) and antibody–drug conjugates (ADCs).

16.19 Xynian Liu - Heidelberg University

Title: Akr1a1a Deficiency Promotes Age-Dependent, Organ-Specific Tissue Alterations in Zebrafish

Akr1a1a is involved in the detoxification of reactive carbonyl species, including acrolein, but its contribution to age-related tissue changes remains unclear. To address this, we generated akr1a1a-deficient zebrafish and examined juvenile, middle-aged, and aged animals.

Akr1a1a deficiency was associated with an age-dependent increase in DNA damage across multiple tissues, including liver, kidney, and muscle, consistent with progressive cellular stress during aging. In addition, mutant zebrafish developed distinct organ-specific phenotypes. In the retina, akr1a1a deficiency was associated with vascular abnormalities, including increased branching, sprouting, and network disorganization. In the kidney, mutants showed age-dependent structural changes, with an early increase in glomerular size followed by a reduction at older ages, suggesting a shift from early adaptation to later structural decline.

Together, these findings identify Akr1a1a as a modifier of age-dependent tissue integrity in zebrafish and indicate that its loss unmasks distinct vascular and renal vulnerabilities during aging. These data support the idea that impaired carbonyl detoxification may contribute to progressive, organ-specific tissue deterioration.

16.20 Diego Andrade-Brito - COS, University of Heidelberg

Title: Molecular-scale analysis of the cellular behavior and plasticity of donor and host cells revealed by inter-species chimeras in fish

Embryonic development reflects an interplay of diverse intrinsic and extrinsic factors that shape cell fate decisions and tissue formation. To disentangle the intrinsic role of developmental modules at the cellular level and to discover the effect of their interaction during embryogenesis, we performed inter-species blastomere transplants in zebrafish (Danio rerio) and medaka (Oryzias latipes). Building on our previous findings that this inter-species transplantation strategy has a strong tendency to generate ectopic (zebrafish) retinae in medaka hosts, we sought to uncover mechanisms of donor cell fate preference by combining genetic perturbations with a single-cell transcriptomic readout. By perturbing rx3, a transcription factor required for retinal cell fate choice, we assessed alternative differentiation paths for zebrafish donor cells. To overcome the challenge of visual readouts for cell fate choice, we profiled whole chimeric embryos and to account for embryo-specific variability, we performed embryo-resolved single-cell RNA-seq on 20 chimeras, coupled to imaging readouts. Our analyses demonstrate a wide-ranging differentiation potential of donor cells, and we characterize similarities and differences of zebrafish and medaka retinal development in these diverse extrinsic contexts. By combining molecular and cellular readouts of how cells develop in diverse external contexts, we shed light on how the cell-intrinsic genetic programs are altered by the species-specific and spatial environmental signals. Overall, this study uncovered new avenues about the molecular and cellular basis of development, evolution, and regulation of cellular traits.

16.21 Lin-Sha Kochsiek - COS Heidelberg

Title : racking mesodermal cell populations with endogenous knock-in approaches

The mesoderm gives rise to a wide range of tissues, including muscle, cartilage, blood, and connective tissues, yet tools to specifically label and trace distinct mesodermal populations in zebrafish remain limited. In particular, several key developmental regulators, such as the T-box transcription factor Ta (tbxta) and the notochord homeobox (noto), lack endogenous knock-in (KI) or robust transgenic reporter lines, restricting the ability to resolve lineage dynamics and cell fate decisions in vivo. To address this, we have developed CRISPR/Cas9-mediated KI strategies to generate endogenous fate-mapping reporters for early axial mesodermal and mesodermally derived cell populations in zebrafish. We have established two novel KI lines, KI(tbxta-p2a-CreERT2);tg(ubi:loxP-eGFP-loxP-mCherry) and KI(noto-p2a-CreERT2);tg(ubi:loxP-eGFP-loxP-mCherry), in which a 2A-CreERT2 cassette is inserted into the endogenous tbxta or noto locus under the native promoter, preserving gene function and enabling temporal control of Cre-recombination. This allows us to study I) the axial and posterior mesoderm as defined by tbxta expression, which drives axial elongation and is required for the formation of a broad mesodermal population (including the notochord, tailbud, and somites), and II) more specifically, the notochord domain, which is specified by noto. Together, these endogenous reporter lines will enable us to flexibly study cell fate dynamics of a range of mesodermally-derived populations in vivo. In addition to the biological application, this work focuses on the methodological challenges of CRISPR KI generation in zebrafish. We optimized strategies for targeting transcription factor loci, including reagent dosages and design of the PCR-generated HDR donors with ~30-40 bp homology arms, achieving KI efficiencies of up to 30% for large (>2 kb) knock-in cassettes. Furthermore, we combine single-cell transcriptomic datasets with spatial expression information from in situ hybridization resources to identify candidate genes for lineage-specific labeling, establishing a pipeline for generating new developmental reporter lines. Together, this work provides both novel mesodermal fate-mapping tools and practical KI approaches applicable to developmental and evolutionary biology research.

16.22 Danila Voronov - Leibniz Institute on Aging

Title: Shared molecular signatures between diapause II and aging in killifish Nothobranchuis furzeri suggest aging during diapause II

Nothobranchius furzeri (Cyprinodontiformes) live in seasonal freshwater ponds that are dried out for most of the year. To survive this, these fish can arrest their embryonic development via diapause. It can occur at three developmental stages: during gastrulation (diapause I), at mid-somitogenesis (diapause II) and before hatching (diapause III). Diapause II (DII) state can last up to multiple years. Diapause is a facultative developmental trajectory, and its alternative trajectory is direct development (DD). Time spent in DII does not affect the lifespan of the adult fish after hatching indicating that either the embryos do not age during diapause or the aging signature is erased post diapause. Despite being a state of arrest, DII nonetheless exhibits dynamic regulation of many genes, a significant proportion of which are regulated during aging as well. This leads to the hypothesis that gene regulation patterns selected to enable diapause could have a pleiotropic detrimental effect in adult life reducing lifespan. We leveraged the next generation sequencing (NGS) technologies to generate multiomic transcriptomic RNA-seq and methylomic RRBS datasets spanning from blastula (3-day post fertilization) to sexually mature (3-week post hatching) fish. Using RNA-seq we looked at DEGs resulting from comparison of DII with DD and genes that follow monotonic trajectories after hatching. We used DNA methylation RRBS data to identify DNA regions differentially methylated in DII and to update the methylomic aging clock model to predict the epigenetic age of DII and DD samples. RNA-seq suggests that gene expression in DII is similar to gene expression during maturation and aging. Aging clocks models show that DII samples exhibit higher epigenetic age than DD samples. Our findings point not only to the shared molecular signatures between DII, maturation and aging, but to embryos exhibiting higher molecular age during DII, which is then reversed upon exit from DII.

16.23 Asalbanoo Farahvashi - Universitätsklinikum Schleswig-Holstein

Title: Zebrafish as an efficient personalised disease model for PRDM16-related cardiomyopathy

Zebrafish have emerged as an efficient and versatile model for investigating genetic cardiomyopathies due to their optical transparency, rapid development, and conserved cardiac physiology. Here, we explore the role of PRDM16 in genetic cardiomyopathy, utilizing zebrafish to functionally test patient-specific genetic variants and to determine their pathogenic potential. Molecular cytogenetic analysis identified four novel PRDM16 variants in individuals with arrhythmogenic right ventricular cardiomyopathy (ARVC) and a truncated variant in a case of left ventricular non-compaction (LVNC). Heart-specific overexpression of these variants in zebrafish exhibited significant changes in structural and functional phenotypic traits including reduced ventricular size and impaired cardiac output, all hallmarks of cardiomyopathic abnormalities. Notably, adult zebrafish hearts expressing PRDM16-D628N and PRDM16-S1059L variants showed increased lipid droplet accumulation, mirroring pathological features observed in patients with ARVC. These findings underscore the potential of zebrafish as a personalized in vivo platform to model PRDM16-related cardiomyopathies, providing insight into disease mechanisms and enabling future therapeutic screening.

16.24 Romane Vanhaeren - University of Liege, Belgium

Title: In Vivo Modeling of MRKH Syndrome Using nr6a1a/b Mutant Zebrafish

The Mayer-Rokitansky-Küster-Hauser (MRKH) syndrome is a rare disorder affecting approximately 1 in 4500 women, characterized by uterine and vaginal aplasia, often associated with kidney and skeletal defects. While TBX6 and GREB1L mutations cause similar anomalies, many MRKH patients lack mutations in these genes, suggesting the involvement of others. Exome sequencing identified two highly pathogenic variants in NR6A1, encoding an orphan nuclear receptor. AlphaFold predictions suggest these mutations impair DNA binding or destabilize its structure. Gel shift assays revealed that mutant NR6A1 proteins have a reduced DNA-binding capacity compared to the wild-type protein.

To explore the link between these variants and the disease, a zebrafish model was generated by inactivating the orthologous genes nr6a1a and nr6a1b. Phenotypic analysis of the nr6a1a mutants revealed reduced body and trunk size, fewer thoracic vertebrae, and absence of the anal fin. In these mutants, the mesonephros displayed altered morphology and reduced size, while pronephric segments and cloacal regions were disorganized. Double nr6a1a;nr6a1b mutants were more severely affected, showing unilateral or bilateral pectoral fin loss, posterior pharyngeal arch defects, and early lethality (12-15 dpf). Gene expression analysis through in situ hybridization revealed altered expression of posterior hox genes, which are crucial for genital tract and kidney development. These findings suggest that NR6A1 mutations contribute to MRKH syndrome, partly through hox dysregulation.

To further investigate disease mechanisms, RNA-seq was performed on WT and mutant zebrafish embryos at the 20-somite stage, confirming the upregulation of posterior hox genes and the dysregulation of many other genes. In situ hybridization (ISH) experiments are ongoing to validate the alteration of these genes. Future studies will include CUT&TAG sequencing to identify genomic loci directly regulated by nr6a1a, shedding light on its transcriptional network.

16.25 Colin Lolos - GIGA Liège, Belgium

Title: Single-Cell Cross-Species Profiling identifies Conserved Transcriptional Networks in Early Pancreatic Tumourigenesis

Background

Pancreatic ductal adenocarcinoma (PDAC) is the most common form of pancreatic cancer and has the worst prognosis among all cancers, largely because it is often diagnosed at advanced metastatic stages. Identifying robust markers of preinvasive disease and elucidating the molecular networks that drive progression from early lesions to invasive carcinoma are therefore critical to improve early detection.

Methods

We generated a zebrafish model in which oncogenic KRASG12D is specifically expressed in pancreatic acinar cells, inducing acinar-to-ductal metaplasia that faithfully recapitulates key features of mammalian pancreatic tumourigenesis. Single-cell RNA sequencing was performed to characterize transcriptional changes occurring at early disease stages. Cross-species comparisons were conducted with published mouse and human pancreatic single-cell transcriptomic datasets to identify evolutionarily conserved genes and signalling pathways associated with pancreatic tumourigenesis. Monocle3 analysis was used to precisely reconstruct the trajectory of tumour cells from normal acinar to malignant states and active regulatory networks along this trajectory were inferred using SCENIC.

Results

Cross-species analyses revealed a striking conservation of genes upregulated during metaplasia, activating common signalling pathways and regulatory programs in zebrafish, mouse, and human. Notably, metaplastic cells reactivate a broad set of developmental genes normally expressed in multipotent pancreatic progenitors. High concordance across species is also observed when reconstructing tumour cell trajectories from acinar to cancerous states, revealing shared gene expression changes along this progression.

Notably, these analyses reveal a set of cytoskeletal and migration-related genes specifically upregulated at the metaplasia–cancer transition, likely conferring invasive potential to these cells. SCENIC analysis further identified regulatory networks that become progressively activated during malignant transformation, suggesting their involvement in the acquisition of cancer-associated traits.

Conclusions

These findings demonstrate strong evolutionary conservation of molecular mechanisms driving pancreatic cancer progression and identify key pathways and regulatory programs that represent promising targets for preventing PDAC development and progression.

16.26 Javier Marin Vasquez - COS, University of Heidelberg

Title: Cell autonomy, plasticity and patterning evolution: fish ionocytes as an experimental paradigm

Conserved cell types can vary dramatically in size, proportion, and distribution across species. In organs hosting multiple cell lineages, their precise coordination is essential for homeostasis and proper post-embryonic growth. It remains unclear, however, the extent of cell types that follow autonomous developmental programs, those that rely on extrinsic signals, and how this rationale varies across species.

We address this question by studying skin ionocytes in different fish species of the Oryzias group. Ionocytes are specialized cells that regulate osmotic balance by pumping ions in and out the cell in response to external salinity. These cells are scattered through the epidermis in a characteristic “salt-and-pepper” pattern and undergo morphological changes depending on environmental conditions. The distribution, morphology and response to environmental cues exhibit a broad phenotypic range among Oryzias species, making them an excellent system that combines several degrees of complexity to study cell autonomous traits.

By combining cell lineage tracing experiments, cell morphology analysis and comparison of inter-species chimeras, we aimed to investigate the embryonic origin of these cells and their degree of cell autonomy in specific traits such as density, distribution and response to external salinity conditions. Our results indicate that, in some Oryzias species, ionocytes follow cell-autonomous programs, while in others they adapt to the physiological environment imposed by the host. These findings might reveal that even highly specialized, conserved cell types can follow different developmental strategies across closely-related species, providing insight into how cellular coordination mechanisms evolve.

16.27 Xinyue Wang - TU Braunschweig

Title: Autophagosome-lysosome fusion impairment triggers secretory autophagy in Spinocerebellar Ataxia Type 13 disease models

Spinocerebellar ataxia Type 13 (SCA13) is an autosomal dominant neurodegenerative disease caused by mutations in the KCNC3 gene. It encodes the voltage-gated potassium channel, which exhibits predominant expression in the Purkinje Cells (PCs), the sole output neurons in cerebellum. The SCA13-causing R420H mutation results in a non-functional transmembrane subunit of KCNC3, which leads to an adult-onset spinocerebellar atrophy. However, the precise correlation between the dysfunctional channel protein and neuronal cell death remains poorly elucidated.
We have genetically modelled SCA13 in zebrafish and cell culture by expressing zebrafish kcnc3R335H or the corresponding human KCNC3R420H pathogenic variants. Affected zebrafish PCs display a progressive degeneration of these key neurons, which causes atrophy of the zebrafish cerebellum like in human. Because the Kcnc3 knock-out mouse model failed to show signs of PC death, the loss of electrophysiological properties of the channel protein alone cannot explain the neurodegenerative phenotype in SCA13. Instead, we have observed a misregulation of degradative autophagy particularly at the stage of autophagosome and lysosome (A-L) fusion. This defect was consistently present in both zebrafish PCs in vivo and cultured cells in vitro in the context of SCA13. As reported for late autophagy inhibition, we therefore investigate whether secretory autophagy is activated as an alternative route to remove cellular content targeted for degradation. Indeed, studies in HEK293-SCA13 cell model showed clear evidence of secretion of autophagic marker in conditioned media suggesting that KCNC3R420H-expressing cells switch from degradative to secretory autophagy in response to impaired autophagosome maturation.

Currently, we aim to elucidate the molecular pathway that is activated to mediate secretory autophagy in our SCA13 models, which is thought to compensate for the compromised degradative pathway. Moreover, we aim to investigate if cargo deployed by secretory autophagy to the extracellular space mediates activation of microglia and triggers inflammation. Such increased levels of neuroinflammation could either serve to relieve cells from harmful content or further damage SCA13-affected PCs by secreting cytokines and cellular stress enhancing metabolites.
Our findings in cell and zebrafish models indicate a close correlation between impaired degradative autophagy and neurodegeneration. Further analysis of novel secretory autophagy pathways will improve our understanding of the underlying SCA13 pathogenesis driving this neurodegenerative disease.

16.28 Melissa Comstock - TU Braunschweig

Title: Corticotropin Releasing Hormone signaling in the larval zebrafish cerebellum

Corticotropin releasing hormone (CRH) and associated proteins are well known for their central role in the mammalian Hypothalamic-Pituitary-Adrenal (HPA) axis regulating curiosity, stress, and anxiety. Several cerebellum-specific disease models in zebrafish with impaired Purkinje or granule cell functions – the key neurons of the cerebellar cortex – have displayed reduced curiosity and increased anxiety as major behavioral signs. In addition, expression analysis in mice have implicated the cerebellum in CRH signaling transduction. However, these signaling proteins are less well understood in zebrafish. We therefore aim to elucidate a potential link between impaired cerebellar function and Crh signaling in zebrafish.
As a basis for further investigations, we have performed a comprehensive mRNA expression analysis of crh ligands and receptors using the Hybridization Chain Reaction (HCR) method. In larvae ranging from 2 to 5 dpf this was performed for the primary ligands, crha, crhb, and urotensin1 (uts1), the two receptors, crhr1 and crhr2, and the crh binding protein (crhbp). We confirm the previous finding that crha mRNA is expressed specifically in the hypothalamus with expression beginning at 3 dpf. mRNAs of the ligands crhb, uts1, and crhbp as well as their receptors crhr1 and crhr2 are observed from 2 dpf and are expressed in discrete cell populations throughout the larval brain. Interestingly, within the cerebellum the two crh receptors are prominently expressed but appear to be localized to different cellular populations. Colocalization experiments using individual transgenic fish lines marking Granule cells (GCs), Purkinje cells (PCs), Eurydendroid cells (ECs), and Bergmann glia were performed. These expression studies showed that mRNA of crhr1 is specifically expressed in cerebellar GCs, while crhr2 mRNA expression is found in PCs and ECs.

As crhr1 appears to be expressed in a single cell type within the cerebellum, whereas crhr2 is expressed in multiple cerebellar cell types, we decided to focus on analyzing the developmental functions of Crhr1 in differentiating zebrafish cerebellar GCs. To genetically target this cell type, we have identified a regulatory element of the granule cell specific atonal1c homolog which served to establish a stable transgenic zebrafish strain Tg(atoh1c:mClover) for monitoring differentiating cerebellar granule cells in vivo. Fluorescent protein expression is initiated in GCs at ~ 2.5 dpf. By 5 dpf expression is noted in the cerebellar GCs and along the rostral midline of the optic tectum. GC-specific expression of mClover was confirmed by HCR coexpression analysis with the GC marker gabra6b.
To investigate the cell-autonomous functions of Crhr1 in individual GCs we have established constitutively active and dominant negative variants of this receptor based on previous investigations. Currently the influence of these variants on cAMP-signaling are investigated. We next aim to express these variants in cerebellar GCs to reveal the consequences of gain- and loss of function of Crhr1 on GC development, morphology and zebrafish behavior. These investigations aim to reveal a better understanding of the importance of CRH-signaling for socio-emotional behavior involving the cerebellum in vertebrates.

16.29 Vanessa Fee Saalmann - UFZ Leipzig

Title: Functional assessment of biomarkers for disruption of the retinoic acid signaling pathway in zebrafish embryos using CRISPR-based knockdown of nuclear receptors

Numerous anthropogenic chemicals continuously enter aquatic ecosystems, where
they can cause adverse effects in organisms. Endocrine-active substances are particularly relevant, as they interact with hormonal signaling pathways and can influence key processes such as growth, reproduction, immune response, and development. However, regulatory assessment currently focuses primarily on the established EATS modalities (estrogenic, androgenic, thyroidal, and steroidogenic mechanisms of action). Other endocrine axes have received significantly less attention, although they also represent sensitive targets for environmental chemicals.
In particular, for the highly conserved retinoic acid (RA) signaling pathway, standardized and validated methods for identifying potentially disruptive substances are largely lacking, while numerous studies point to adverse effects associated with disruption of RA signaling in environmental organisms.
As part of the RetiNAM project, potentially specific biomarkers (morphological, behavioral, and gene expression changes) were identified in zebrafish embryos through exposure to model chemicals. RetiNAM focused on the binding of these chemicals to the retinoic acid receptor (RAR) or retinoid X receptor (RXR) as the molecular initiating event. These previously identified biomarkers are being investigated in this study using a CRISPR/Cas9-based knockdown approach.
CRISPR/Cas9 knockdown is performed at the single-cell stage of the embryos via automated microinjection. This involves the use of guide RNAs (gRNAs), which are designed to guide Cas9 to the target gene and induce site-specific DNA cleavage.

The study is conducted transiently, i.e., the embryos are examined for morphological at day 4 post-fertilization. To systematically detect developmental changes, a key focus is on the automated image-based quantitative phenotyping of the entire embryo.
It is hypothesized that both chemical-mediated agonism or antagonism of RAR and RXR, as well as the knockdown of these receptors, induce specific phenotypic
changes, thereby enabling the identification of RA signaling pathway-specific effects and assessment of the relevance of previously identified potential biomarkers for disruption of RA signaling.

16.30 Arish Shah - COS, University of Heidelberg

Title: Single-cell characterization of cell fate decisions in a ribosomopathy model

Ribosome biogenesis is essential for all proliferative cells, yet mutations in ribosome production factors often produce strikingly tissue-specific developmental phenotypes. In vertebrates, craniofacial structures are particularly sensitive to disruptions in ribosome biogenesis, but the mechanisms underlying this lineage specificity remain unclear. Using zebrafish mutants we investigate how impaired rRNA transcription impacts craniofacial cartilage development.

Embryos carrying mutated alleles of polr1a, the catalytic subunit of RNA Pol I, exhibit severe craniofacial defects, failing to generate both cranial neural crest cell (cNCC)-derived and mesoderm-derived cartilage elements. Removal of tp53 restores overall morphological growth and rescues mesoderm-derived cartilage structures, demonstrating that a component of the growth defect is mediated by canonical cell cycle impairment and apoptosis. Unexpectedly, cNCC-derived cartilage fails to recover in the polr1a; tp53 double mutant background, revealing a tp53-independent requirement for Pol I activity during cNCC differentiation into cartilage.

To resolve lineage-specific responses at cellular resolution, we performed single-cell RNA sequencing on polr1a CRISPant embryos. While global activation of the tp53 and integrated stress responses is observed across lineages, cNCC-derived mesenchyme uniquely exhibits disruption of differentiation programs. These findings indicate that neural crest cells are not merely dying, but are failing to execute lineage-specific differentiation under conditions of rRNA insufficiency.

Consistent with Pol I's role in nucleolar assembly, polr1a mutants display altered nucleolar architecture, suggesting that disruption of nucleolar organization – rather than simple loss of translational output – may underlie the lineage-specific phenotype.

We propose that impaired nucleolar integrity, defective ribosomal protein incorporation, and consequent proteotoxic stress may contribute to neural crest-specific vulnerability. Together, this work provides a mechanistic framework for understanding the tissue specificity of ribosomopathies and supports a model in which ribosome biogenesis governs cell fate decisions not only through translational capacity, but also through maintenance of nucleolar structure and proteostasis.

16.31 Quincy Van den Bosch - RWTH Aachen

Title: Accelerated Early Vascular Aging in the Turquoise Killifish: From Morphological Remodeling to Degenerative Mineralization

Background: Advanced age is a primary independent risk factor for atherosclerosis, hallmarked by chronic, systemic inflammation and dysfunctional immune responses that drive cumulative cardiovascular damage. These processes can manifest as endothelial dysfunction, immune infiltration, calcification, and plaque formation. However, studying the transition from natural aging to vascular pathology is hindered by the long lifespans of traditional models. We propose the Turquoise Killifish as a unique vertebrate model to investigate the etiology of vascular aging in a compressed timeframe due to its rapid transition from adulthood (~6-8 weeks) to aged (~20 weeks).

Methods: We utilized the bulbus arteriosus, the anatomical equivalent of the aorta in killifish as a high-speed model for vascular remodeling. Longitudinal analysis was conducted at 7, 23, 37, and 43 weeks (n= 32, 4males/4 females per group). Killifish were fed standard diet (bloodworms, 0.021g fat/kcal) according to standard killifish husbandry. Morphological and pathological transitions were quantified using Sirius Red and Alizarin Red histology to map matrix remodeling.

Results: Analysis reveals a distinct biphasic progression of vascular remodeling. By week 23, the bulbus arteriosus exhibits significant aging symptoms, including increased tunica media thickness($p=0.015$) and declining cell density($p=<0.001$). While these parameters plateau after 23 weeks, structural integrity continues to decline characterized by progressive remodeling. Sirius Red staining identified collagen-rich deposits that expand($p=0.018$), indicating chronic fibrotic maturation. Concurrently, we observed a progressive increase of medial mineral deposits from 23 weeks onwards ($p=0.014$), suggesting a transition from active remodeling to a passive, degenerative mineral-layering process.

Conclusion: Integrating histological evidence of rapid mineralization and fibrosis, this work positions the killifish as a premier, high-throughput model for exploring the drivers of age-related vascular stiffness. Future directions include single-nucleus RNA sequencing (snRNA-seq) to resolve the underlying cellular dynamics and signaling pathways responsible for this rapid remodeling.

16.32 Mohammed Lukman Rafi - Justus Liebig University

Title: Interleukin-11 as a Regulator of Redox Signaling During Zebrafish Tissue Regeneration

The zebrafish heart possesses the remarkable capacity for scar-free regeneration following tissue loss or damage, providing critical insights into how to combat fibrosis and scarring, which is the outcome of tissue damage in mammals. We had identified il11 signaling via its cognate receptor il11ra is simultaneously pro-regenerative and prevents mammalian like scarring as evident from our il11ra-/- mutant animal model which don't regenerate multiple tissue types. We are now interested in figuring out the downstream mechanism that il11 is instructive for successful regeneration and what are the major physiological hindrance upon loss of il11 signaling during failed regeneration. One such physiological process is redox regulation, indeed modulated for successful regenerative outgrowth as evident form literature to elicit early injury response and inflammation. Our sc-RNA seq data from the il11ra-/- stromal cell population hint of various redox regulated and related gene markers dysregulated during the vital window of regeneration. We observed in the mutants that components of the thioredoxin (txn)-peroxiredoxins(prdx) are significantly downregulated in the mutants reasoning ROS or H2O2 dysregulation. We profiled genes associated with this antioxidant pathway using qPCR and saw significant downregulation of key genes such as txna and prdx6. We also observed by using live larval zebrafish imaging with fluorophores specific for ROS and H2O2 that the mutants showed decreased staining at least by 3 folds relative to the WT confirming the role of a dysregulated redox homeostasis in il11ra-/- mutants. Putative cell proliferative, dedifferentiation mechanism that are reliant on this redox regulation are our downstream physiology of interest. How il11 regulates such a pleotropic and fast acting component of injury during early timepoint's to successfully drive the tissue to a pro-regenerative program is the question we are interested in addressing. Taken together our study will try to delineate the role for il11 in modulating redox signaling in the context of injury response which will give a clarity on how there is a role for regeneration.

16.33 Bernd Gahr - University Hospital Ulm

Title: Loss of Traf7 attenuates the proliferative capacity of cardiomyocytes

Despite cardiovascular diseases remaining the leading cause of mortality worldwide, the molecular mechanisms that regulate cardiomyocyte homeostasis and proliferation remain incompletely understood. A deeper understanding of the fine-tuned control of cardiomyocyte proliferation is essential for elucidating the molecular processes underlying normal cardiac development and susceptibility to cardiovascular disease.

Here, we describe the recessive embryonic-lethal zebrafish mutant herzbuckel, which exhibits impaired cardiac growth as a result of reduced cardiomyocyte proliferation. Positional cloning identified a loss-of-function mutation in the zebrafish tumor necrosis factor receptor associated factor 7 (traf7) gene. The mutation introduces a premature stop codon, resulting in truncation of the Traf7 protein at amino acid position 519. To confirm traf7 as the causative gene, we performed both Morpholino-mediated knockdown and CRISPR/Cas9-mediated knockout experiments. Both experiments recapitulate the herzbuckel phenotype. Finally, traf7 mRNA injection was able to rescue cardiomyocyte numbers in the developing zebrafish heart.

To determine whether the role of Traf7 in cardiomyocyte proliferation is evolutionarily conserved, we analyzed the effects of TRAF7 knockdown in neonatal rat cardiomyocytes using AAV-mediated shRNA delivery. Similar to our observations in the developing zebrafish heart, TRAF7 depletion resulted in a significant reduction in cardiomyocyte proliferation. These findings suggest that the role of Traf7 in regulating cardiomyocyte proliferation is conserved across vertebrates.

In the context of cellular proliferation, the E3 ubiquitin ligase TRAF7 has been implicated in both the activation and repression of NF-κB signaling. NF-κB is a key regulatory pathway involved in diverse cellular processes, including cell cycle progression and autophagy. We found that the loss of TRAF7 in rat cardiomyocytes alters the phosphorylation status of NF-κB, suggesting impaired pathway regulation.

Collectively, our findings support an important and evolutionarily conserved role for TRAF7 in the regulation of cardiomyocyte proliferation.

16.34 Sabrina Afsa - University of Messina, Italy

Title: Polystyrene Microplastics (PS-MPs) May Modulate Appetite Regulation and Metabolic Homeostasis in Zebrafish under different feeding conditions: Could These Alterations Contribute to an Obesogenic Response?

Environmental contamination by microplastics (MPs) is well documented; however, their obesogenic potential and associated metabolic effects remain unclear and warrant a particular attention. Therefore, this study aimed to assess the effects of polystyrene microplastics (PS-MPs) on appetite-related biomarkers and metabolite profile in zebrafish under normal feeding and overfeeding conditions.

To this aim, adult zebrafish were divided into four groups including (1) normally fed group, (2) normally fed group exposed to PS-MPs, (3) overfed group, (4) overfed group exposed to PS-MPs. Zebrafish were exposed to 3-µm PS-MPs (200 µg/L) for four weeks. At the end of the experiment, immunofluorescence was performed on whole zebrafish sections, using confocal laser microscopy, to study appetite-related biomarkers in different organs. Proton Nuclear Magnetic Resonance (1H NMR)-based metabolomics was also applied to assess the metabolic alterations in zebrafish intestine.

The obtained results showed an altered immunoreactivity for both leptin and orexin in the intestine, liver and brain across all experimental conditions, when compared to control. Particularly, an impaired anorexigenic signaling was found within overfed group and PS-MPs-exposed groups compared to normally fed group, indicating a possible modulation in both peripheral and central appetite signaling, particularly under combined nutritional and environmental stress. On the other hand, 1H NMR-based metabolomics revealed a general reduction of metabolites (mM) involved in aerobic (glucose, pyruvate and succinate) and anaerobic (lactate) energy metabolism in zebrafish intestine across all groups with respect to control, particularly in the overfed group exposed to PS-MPs. Furthermore, significant reductions in cholate and urocanate were also detected, suggesting an impairment in bile acid and amino acid metabolism, respectively.

Overall, these findings suggest that PS-MPs exposure, under normal feeding or overfeeding, may alter appetite signaling and metabolic pathways, probably underlying obesogenic effects. However, further analyses are ongoing to complement these preliminary results.

16.35 Marta Bošnjaković - Ruđer Bošković Institute

Title: Promoter-level annotation of the adult zebrafish regulatory landscape using CAGE-seq

Zebrafish (Danio rerio) is a well-established vertebrate model organism widely used in developmental biology, molecular genetics, cancer research, toxicology, drug discovery, and regenerative medicine. Its experimental accessibility and conservation of many genes and regulatory pathways with other vertebrates, including humans, make it a valuable system for both fundamental and biomedical research. A comprehensive understanding of its regulatory genome is therefore important for interpreting gene expression and regulatory mechanisms across these research areas. Despite extensive genomic resources, however, the transcription initiation landscape of adult zebrafish tissues remains underexplored. The DANIO-CODE initiative has substantially advanced zebrafish genomics by providing high-resolution maps of transcription initiation, but with most efforts focused on embryonic development. Here, we extend this resource to adult tissues using Cap Analysis of Gene Expression sequencing (CAGE-seq). We provide promoter annotations across seven adult zebrafish organs: brain, heart, skin, liver, gastrointestinal tract, testes, and ovaries. We identified 15.348 active promoters, providing base-pair resolution transcription start site (TSS) maps that complement and validate existing epigenomic datasets (e.g. ChIP-seq and ATAC-seq). This resource will refine the annotation of chromatin-defined regulatory elements, enhancing the interpretation of open chromatin and histone modification signals. This work enhances promoter annotation, making adult zebrafish research more accurate and precise for biomedical applications. Overall, our dataset supports better functional studies, such as CRISPR guide design and transcript-level expression analysis and reveals both shared and tissue-specific promoter activity in adult zebrafish.

16.36 Jonas Baumann - HIPS Saarbrücken

Title: Multi-Organ Toxicity Screening of Drug Candidates Using Transgenic Zebrafish

The discovery and development of new drug candidates still require in vivo testing in approved animal studies. According to EU Directive 2010/63/EU, zebrafish (ZF) embryos and larvae before 120 hours post-fertilization (hpf) are not covered by the regulatory framework governing animal experimentation and therefore do not have protected status. In recent years, the ZF embryo model has gained increasing attention as an alternative approach for the early screening of new drug candidates prior to potential testing in regulated animal studies.

ZF embryos develop major organ systems as early as 48–72 hpf, making them a valuable model for investigating organ-specific toxic effects of compounds. A wide range of transgenic ZF lines is available that express fluorescent reporter proteins in specific organs or cell types. At HIPS, three such lines are used to develop a multi-organ toxicity screening approach for the detection of renal, hepatic, and ototoxic effects of drug candidates: Tg(wt1b), Tg(fabp10a), and Tg(pou4f3).

This poster presents the experimental procedures and workflows used to develop and apply these transgenic ZF-based screens for semi-automated multi-organ toxicity assessment. The approach enables the detection and assessment of compound-induced toxic effects across different organs using complementary transgenic zebrafish lines, supporting the early evaluation of potential organotoxicity during drug development.

16.37 Alba Madrero-Pardo - Ruđer Bošković Institute

Title: Mapping non-coding functional elements in allotetraploid Cyprinus carpio embryo development reveals subgenome variation of transcription regulation

Whole-genome duplication is a major driver of vertebrate evolution, yet how duplicated regulatory genomes diverge and re-coordinate over developmental time remains poorly resolved. Common carp (Cyprinus carpio), an allotetraploid cyprinid and key comparative model for zebrafish, offers a natural system to study this process, as its two chromosomally distinct subgenomes have evolved separately since hybridisation (11-13Mya) while sharing a common developmental program. We generated a developmental regulatory atlas across 12 stages, combining transcriptome dynamics (RNA-seq) with chromatin accessibility (ATAC-seq), transcription start site mapping (CAGE-seq), and histone modification profiles (ChIP-seq) to trace subgenome regulatory evolution through embryonic development. Across 14.849 one-to-one homeolog pairs, expression asymmetry peaks around the maternal-to-zygotic transition, with subgenome B predominating, and resolves into increasing coordination through development. Comparing 254.276 annotated regulatory elements between subgenomes, we find regulatory divergence is highest in early development and converges toward the phylotypic period, a pattern driven specifically by enhancers. At the same time, promoters retain a stable evolutionary bias. This extends the hourglass model of developmental constraint to the subgenomes of an allotetraploid. Subgenome B retains significantly subgenome-specific enhancers, while subgenome A shifts toward homeologous enhancers by the phylotypic stage, revealing directional, asymmetric regulatory evolution between subgenomes. Comparison with zebrafish shows concordance between sequence conservation and chromatin detection of regulatory elements, with subgenome B retaining more elements resembling this ancestral, zebrafish-like regulatory state. Together, these results reveal how developmental constraint and regulatory innovation jointly shape genome evolution following the fourth round of vertebrate whole-genome duplication.

16.38 Sophia Levitskaja - Max Dellbrück Center Berlin

Title: Integrating spatial transcriptomics and live imaging to resolve extrinsic drivers of cranial neural crest fate

The cranial neural crest (CNC) is a migratory, multipotent embryonic cell population that gives rise to remarkably diverse cell fates. As CNC cells migrate, they continuously interact with the surrounding tissue environment, raising the question of which extrinsic signals, and to what extent, shape their fate decisions. Addressing this requires jointly profiling a cell's molecular identity, its migratory trajectory, and its interactions with the environment over time.

We aim to integrate spatial and single cell transcriptomics with live imaging to link individual cell trajectories to their eventual fates, and to identify extrinsic signaling interactions in space and time. We use spatial transcriptomics (openST) reconstructed in 3D and single cell RNA sequencing to profile both CNC cells and their surrounding environment tissue in parallel, alongside live imaging of CNC cell behavior.

To date, we have generated and annotated scRNA-seq time courses for both CNC and environment cell populations, and acquired preliminary 3D openST spatial data. This establishes the foundation for the next phase: systematically identifying extrinsic signaling interactions that correlate with specific CNC fate outcomes, and using live imaging to link individual cell trajectories directly to their eventual fate.

Posters Session 2

Thursday 17th, 2026

17.10 - 19.00

17.01 Julian Rebentisch - COS, University of Heidelberg

Title: Investigating the gene regulatory code of zebrafish craniofacial cartilage development with endogenous protein tagging and deep learning

Gene regulation is orchestrated by the fine-grained interplay of transcription factors, promoters and enhancers. During zebrafish development, precise gene regulatory control is essential to achieve the coordinated migration, differentiation and morphogenesis of chondrocytes that form the early skull. In the posterior skull, two separate embryonic lineages, neural crest and mesoderm, generate adjacent but distinct cranial elements. Previous work from the lab has demonstrated that despite a common cell type, the transcriptional profiles of these populations reflect their lineage origins, suggesting that the gene regulatory mechanisms driving expression of the core chondrocyte regulatory module, sox5/6/9a, may be lineage-specific. To identify lineage-specific chondrocyte enhancers, we are using cleavage under targets and tagmentation (CUT&Tag). To overcome the shortage of validated antibodies, we introduced small epitope-tags to the endogenous protein coding sequences of key chondrocyte transcription factors. As a complementary approach, we are leveraging sequence-to-function models to gain a mechanistic understanding of the craniofacial gene regulatory code at nucleotide resolution. We will leverage the published DeepZebraFish model to understand regulatory sequences active in craniofacial cell types. Through the use of in silico evolution guided by the deep learning model, we aim to generate synthetic enhancer sequences capable of driving cell-type specific expression. We will validate these sequences in vivo by coupling them to reporter gene expression. Together, this project will advance our understanding of active enhancer sequences in early craniofacial development and identify lineage-specific gene regulatory drivers of chondrocyte differentation.

17.02 Zixuan Huo - COS, University of Heidelberg

Title: Decoding craniofacial structure variation and genetic dependencies using the Medaka Inbred Kiyosu-Karlsruhe Panel

The craniofacial complex is a key vertebrate trait, serving as the structural basis of predation and anterior organ positioning and protection. Among vertebrate species, craniofacial structures undergo significant morphological adaptation, underlying the substantial variability seen within and across species. Understanding the link between the genetic control and phenotypic outcome of this complex tissue is a fundamental question in evolutionary and developmental biology. To study genetic mechanisms that contribute to diverse craniofacial phenotypes, I am employing the medaka inbred Kiyosu-Karlsruhe (MIKK) panel, an inbred panel of Medaka (Oryzias latipes) strains that capture a range of genetic and phenotypic variation (Fitzgerald et al. 2022). I am combining genetic mapping of defined morphological phenotypes with scRNA-seq based high-resolution phenotyping of embryonic development to understand the molecular and cellular mechanisms underlying these differences. I characterized the craniofacial diversity within the MIKK panel based on adult phenotypes and subsequently quantified craniofacial morphology at multiple embryonic stages. Based on these analyses, I selected 4 “extreme” lines displaying the most structural variance, taking into account consistency across embryonic stages. To determine whether measurable phenotypes co-segregate with the same extreme genotype and specific genetic changes underlying phenotypic differences, I am using a segregation crossing scheme followed by genetic mapping. Concurrently, I will phenotype “extreme” lines with scRNA-seq on multiple developmental stages to uncover the molecular and cellular changes during early skull development. Together, my goal is to link genetic, cellular and phenotypic aspects of craniofacial morphology.

17.03 Alina Bektimirova - Institute of Experimental Cardiology, Heidelberg

Title: Establishment of a 3D culture system to investigate mechanisms and discover novel molecules that promote cardiomyocyte invasion of fibrotic scar tissue

Myocardial infarction in mammals causes irreversible fibrotic scar due to the inability of cardiomyocytes (CMs) to regenerate. In contrast to mammals, zebrafish myocardium can fully regenerate following apical resection and cryoinjury, an injury model used to mimic myocardial infarction. Our recent study indicates that CM protrusion and invasion into the fibrotic tissue are crucial for zebrafish cardiac regeneration (Constanty, Wu, et al, 2025, Nat. Commun.). This CM invasion is likely regulated by two mechanisms: (i) CM-intrinsic actin cytoskeleton remodeling and subsequent cell invasion and (ii) macrophage-mediated extracellular matrix (ECM) remodeling at the border that enables CM invasion. The molecular and cellular mechanisms of CM invasion, CM:macrophage interaction and the role of ECM remodeling, however, are still largely unknown.

Here, we aim to develop a 3D CM culture system to recapitulate CM invasion in vitro. As a proof-of-concept, we use zebrafish CMs, known for their regenerative capacity in vivo for multiple reasons: we have observed the invasion of CMs into injured tissue using live-imaging, and we have multiple genetic models that inhibit or promote CM protrusion and invasion that we have characterized in vivo. In order to model the post-injury environment in vitro, we used atomic force microscopy (AFM) to measure tissue stiffness across the different regions of regenerating heart (7 days post cryoinjury). To test the invasion of CMs in vitro, we used a hydrogel system modified with cell adhesive peptide RGD and with stiffnesses corresponding to measured values by AFM. The invasion assay revealed progressive migration of CMs over a 3-day culture period. By day 1, most CMs remained at or near the surface of the hydrogel layer. By day 3, numerous CMs had penetrated deeper into the hydrogel, with some reaching close to the bottom of the matrix.

Overall, our hydrogel-based invasion assay indicates that zebrafish CMs migrate in response to matrix stiffness.

Currently, we are optimizing this 3D culture system to investigate the importance of CM-intrinsic mechanisms versus the role of other cell types at the BZ to remodel ECM and enable CM invasion. In the future, applying these insights to human iPSC-derived CMs could guide the development of therapeutic strategies to replace fibrotic scar tissue and restore heart function after myocardial infarction.

17.04 Deung-Dae Park - University of Ulm

Title: Washc4 Maintains Cardiac Cellular Homeostasis and Mitochondrial Function During Aging in Zebrafish

Cardiac aging is characterized by progressive cellular stress and metabolic remodeling, yet upstream pathways linking these processes to age-dependent cardiac remodeling remain incompletely defined. WASHC4 is a subunit of the WASH complex that regulates endosomal actin dynamics and membrane trafficking, but its role in cardiac homeostasis is poorly understood. Here, zebrafish models with partial or complete loss of Washc4 were used to assess age-dependent cardiac phenotypes. Washc4-deficient zebrafish developed normally during embryogenesis and maintained cardiac morphology early in life. With aging, washc4−/− animals exhibited impaired growth, reduced survival, and progressive cardiac remodeling, including ventricular enlargement, epicardial lipid accumulation, and altered cardiomyocyte geometry. Hearts from washc4−/− fish showed induction of senescence and SASP-associated transcripts. Quantitative proteomics revealed coordinated remodeling of mitochondrial energy metabolism pathways, accompanied by altered cardiomyocyte respiration, increased oxidative stress, and DNA damage signatures. Overall, Washc4 deficiency is associated with age-dependent mitochondrial remodeling and senescence-related changes in the heart.

17.05 Caroline Zandecki - KU Leuven, Belgium

Title: A killifish-based in vivo screening platform identifies promising anti-aging compounds

Population aging is rapidly increasing worldwide and is accompanied by a strong rise in age-related disorders, including neurodegenerative diseases, sarcopenia and frailty. Because aging itself is the primary risk factor in many of these chronic conditions, interventions targeting fundamental aging mechanisms may provide broader therapeutic benefits than disease-specific approaches alone. However, the discovery and validation of candidate geroprotective compounds requires robust in vivo screening, capable of assessing functional and molecular aging phenotypes in a time-efficient manner. To address this need, we established a screening platform using the African Turquoise killifish (Nothobranchius furzeri, GRZ-AD), a naturally short-lived vertebrate model with growing recognition in aging research.

Within a consortium, biobased molecules were generated with presumed anti-senescent properties and pre-selected using toxicity screening pipelines. Here, we present the in vivo evaluation of these candidate anti-aging compounds in our adult killifish screening platform.

This study focused on two groups of biobased compounds with distinct biological signatures. A first candidate compound was tested for effects on the aging brain. Preliminary analysis suggests changes in senescent cell load. In parallel, we could identify two derivatives out of a second compound group, showing modulated muscle-related phenotypes during aging. Behavioral analysis using open field assessment revealed indications of changed locomotor performance, while muscle health measurement using electrical impedance myography suggested altered muscle integrity. Furthermore, exploratory qPCR profiling indicated modulation of molecular signatures in skeletal muscle tissue.

Together, these results provide proof-of-concept that our killifish platform can detect functional and molecular responses to novel candidate anti-aging compounds. This screening approach enables rapid evaluation of behavioral, physiological, and molecular aging phenotypes and supports the identification of promising lead compounds for future translational development.

17.06 Laura Herrera Astorga - COS, University of Heidelberg

Title: Assessing retinal regeneration across medaka COS-MIKK panel lines with contrasting nervous system repair capacities

Retinal regeneration capacities vary widely across and within vertebrate groups. Comparative approaches have been valuable to explore the processes shaping the sustained neurogenesis that enables retinal regeneration in many teleosts, but constrain it in mammals. In contrast to zebrafish (Danio rerio), the Cab strain of medaka (Oryzias latipes) shows restricted retinal regeneration capacities. However, analysis of other medaka strains has revealed intra-specific variability of the regenerative capacities of the nervous system. To explore the genetic basis shaping those differential regenerative capacities we aim to analyze retinal regeneration in a unique resource, the medaka COS-MIKK inbred panel. For this, we set up a pipeline quantifying regeneration in a highly reproducible and scalable retinal injury and staining protocol. Retinal regeneration was assesed in hatchlings of five COS-MIKK inbred lines previously shown to display differential spinal cord regenerative capacities. This preselection allows to determine whether the mechanisms associated with regeneration in different sub-structures of the central nervous system rely on shared or independent modules. In the retina, the regenerative/non-regenerative phenotype was assesed by the injury-triggered presence of neurogenic clusters in the inner nuclear layer. Our results indicate a variable regeneration potential across the lines analyzed, not clearly linked to spinal cord regeneration abilities. They also point towards differential retinal regeneration capacities in different medaka inbred lines, highlighting the presence of intra-species and individual regenerative capacities. Additionally, the pipeline developed here, will allow to further characterize the COS-MIKK panel and perform quantitative trait loci (QTL) mapping, in order to identify genes associated with the retinal regenerative capacity and advance our general understanding of the molecular basis of retinal regeneration.

17.07 Daniela Vallone - IBCS-BIP KIt Karlsruhe

Title: Evolution of light-regulated transcriptional control in vertebrates

Fish cell biology is very much shaped by sunlight. Direct light exposure triggers changes in the expression of genes as well as miRNAs that are functionally connected with the circadian clock, DNA damage repair, mitochondrial function and heme metabolism. ROS, MAPK signalling as well as the D-box promoter enhancer element play a central role in directing light inducible gene expression in fish. D-box binding transcription factors belonging to the PARbZip family represent convergence points for regulation by light, UV and ROS. We have documented fundamental changes in this signalling cascade in mammals as well as in species of blind cavefish that have evolved for millions of years in complete isolation from sunlight. Using a comparative study involving zebrafish, blind cavefish and mouse cell lines, we reveal how the PARbZip transcription factor TEF is regulated by ROS and light via phosphorylation by the stress MAP Kinases, jnk and p38. Changes in these phosphorylation sites in cavefish and mammalian TEF account for loss of regulation by ROS and light in these species. Our results reveal that sunlight-driven signalling pathways and gene expression control mechanisms represent key targets for evolutionary adaptation in vertebrates.

17.08 Petra Petrašiūnaitė - EMBL, University of Heidelberg

Title: Exploring the interplay between tempo and reproducibility in embryonic development

Embryonic development is precisely timed and coordinated to ensure a robust outcome in a changing environment. However, a growing amount of evidence suggests that accelerating development by using cues like high temperature increases error rate and leads to a loss of reproducibility. In the present work, I am exploring the relationship between developmental rate and reproducibility of embryogenesis by testing the opposite approach: whether deliberately slowing developmental tempo increases developmental reproducibility.

I test this hypothesis in zebrafish embryos using two complementary approaches: lowering temperature and tuning down translation. Developmental rate can be readily manipulated by lowering temperature; however, temperature shifts also induce stress responses and affect multiple cellular processes, making it difficult to disentangle the effects of developmental slowing from the effects of temperature sensing itself. To overcome this limitation, I use low-dose pharmacological translation inhibition as an alternative strategy to reduce developmental speed while avoiding direct perturbation of temperature-sensing pathways. In order to quantify developmental delay effects, I use a deep learning-based automated staging framework that enables measurement of developmental progression across treatments. My findings suggest that inhibitors with distinct mechanisms of action differ in their ability to delay development, indicating that some parts of the translation process are more tunable for controlling developmental rate than others. I also find that some developmental timepoints appear to be more sensitive to slowing than others.

To investigate the mechanisms underlying these developmental delays and quantify reproducibility across scales, I am integrating the phenotypic measurements with individually-resolved single-cell RNA-seq data from zebrafish embryos developing under translation inhibition or at reduced temperature.

On the molecular level, I will investigate whether the same pathways control the emergence of the delayed development phenotype across treatments. On the cellular level, I will determine whether different cell types react differently to a slower developmental regime, i.e. whether some cell types show higher sensitivity than others as is known to be the case for acceleration. Finally, on the organismal level, I will look into cell type composition changes across embryos in response to a slower developmental regime.

17.09 Anja Bühler - Ulm University Hospital

Title: In Vivo Small Molecule Screen Identifies Calcium Channel Inhibition as a Regulating Cue for Cardiomyocyte Proliferation and Heart Size

During vertebrate heart development and tissue homeostasis, maintaining precise organ size and structural integrity relies heavily on the tightly regulated proliferation of cardiomyocytes (CMs). While adult mammalian CMs lose their proliferative capacity postnatally, adult zebrafish (Danio rerio) maintain a highly plastic cardiac landscape capable of robust CM cell cycle re-entry, dedifferentiation, and complete tissue regeneration. Uncovering the endogenous physiological cues and molecular networks that drive CM proliferation during embryonic cardiogenesis provides crucial insights into how organ growth is controlled and how regenerative plasticity is maintained in the adult fish.

To systematically identify novel regulators of cardiac growth, we performed an in vivo high-throughput screen (HTS) of 485 small bioactive molecules using zebrafish embryos. We identified Cilnidipine, a dual L- and N-type calcium channel blocker, as a potent pro-proliferative hit. Morphometric analysis revealed that embryonic Cilnidipine treatment results in a significantly enlarged ventricle. Crucially, quantification of CMs confirmed this phenotype is driven by an increase in total ventricular CM numbers and elevated proliferation rates, rather than cellular hypertrophy.

To investigate the physiological mechanism linking ion channel activity to cell cycle progression, we conducted in vivo calcium imaging. Treated embryos exhibited severely blunted intracellular Ca^{2+} flux, indicating that the suppression of physiological calcium transients may act as a direct trigger for CM proliferation. Our ongoing efforts aim to dissect the downstream Ca^{2+}-dependent signaling pathways and transcriptional networks mediating this hyperplastic response. Additionally, we are evaluating the evolutionary conservation of this physiological mechanism using human iPSC-derived CM models to understand how calcium dynamics dictate vertebrate CM proliferative capacity. Altogether, these findings point to a compelling correlation between Ca^{2+}-signaling and CM proliferation, establishing a framework to further investigate how intracellular ion dynamics influence proliferative plasticity in the vertebrate heart.

17.10 Essaikiammal Konar - University of South Bohemia, Czech Republic

Title: Postovulatory oocyte aging disrupts early embryonic development and germline formation via oxidative stress in zebrafish

In zebrafish, the embryo relies on maternally deposited factors to regulate cell cycle progression, axis formation, and germline specification prior to zygotic genome activation. The developmental competence of an oocyte thus depends on the integrity of maternal determinants that pattern the earliest cell fate decisions. Although postovulatory oocyte aging (POA) is known to reduce egg quality and developmental competence of embryos, its impact on maternally regulated developmental processes remains poorly understood. Here, we investigated the effect of short-term POA on early embryogenesis, germ plasm dynamics, and primordial germ cell (PGC) formation using the Tg(egfp:ddx4) zebrafish line. Oocytes were aged for 2 hours post-ovulation in vitro prior to fertilization and compared to a freshly ovulated group as controls. Embryos derived from aged oocytes exhibited a significant reduction in PGC number. The embryos from the aged group also showed a higher incidence of mis-migrated PGCs, indicating that POA affects both PGC specification and migration. Consistent with oxidative stress being one of the major drivers of POA, aged oocytes showed a 2.5-fold increase in ROS levels. Exposure of freshly collected oocytes to hydrogen peroxide similarly reduced PGC numbers, supporting a causal role of oxidative stress in germline defects caused by POA. To identify potential mechanisms, we quantified the efficiency of ooplasmic segregation during early embryogenesis. POA significantly impaired ooplasmic transport, a process required for proper redistribution of germ plasm determinants. Given that germ plasm components such as dnd1, dazl, and ddx4 are maternally deposited and require precise cytoplasmic localization, we analysed the localization of these mRNAs by tomography PCR. The distribution of germ plasm associated mRNAs was similar in control and aged group embryos which suggests that POA does not strongly affect germ plasm RNA transport. Together, these findings indicate that POA-induced oxidative stress disrupts cytoskeletal-mediated bulk cytoplasmic movement and PGC formation during early development without affecting microtubule-mediated mRNA transport.

Consequently, oocyte aging compromises germline development and alters developmental patterning. Our study highlights that oocyte quality affects the maternally controlled developmental programs and identifies oxidative stress as a key mechanism linking postovulatory aging to defects in germline formation.

17.11 Aaron Alejandro Torres Martinez - University of South Bohemia, Czech Republic

Title: Morphological and molecular signatures of ovarian aging in zebrafish

Aging is a pivotal topic due to its obvious relevance to human health. Aging studies have been conducted in numerous model organisms; however, most have focused on the aging of somatic organs, while comparatively little attention has been given to gonadal aging. Multiple mechanisms have been implicated in the aging process such a genetic damage, oxidative stress, cell death and microenvironmental alterations. In this study, we used zebrafish to investigate the mechanisms underlying ovarian aging and the decline in fertility. We analyzed the ovaries of 3-year-old (aged) and 6-month-old zebrafish (young) using histology and bulk RNA sequencing to characterize age-associated degenerative processes in the ovary. Young zebrafish ovaries contained abundant secondary growth (SG) oocytes and scattered primary growth (PG) oocytes associated with the germinal epithelium. Germ cell profiles were similar among individuals. In contrast, aged zebrafish ovaries displayed a wide range of germ cell profiles and histological degenerative features, including reduced numbers of oocytes, increased ovarian stroma, and infiltration of immune cells. The most notable alteration was the increased degeneration of PG oocytes ($p<0.01$), leading to the formation of scar-like areas with immune cells infiltration ($p<0.05$). Interestingly, no differences were observed in the atresia rates of SG oocytes ($p=0.97$); however, differences were detected in the percentage of non-atretic SG oocytes ($p<0.01$), which may reflect reduced fecundity in aged females. To explore the molecular basis of ovarian aging, we conducted bulk RNA sequencing to compare the transcriptomic profiles of ovaries from aged and young zebrafish. Global gene expression analysis revealed clear clustering among young ovaries, whereas aged ovaries showed greater dispersion and lacked a distinct clustering pattern. This may reflect differences in the degree and progression of ovarian aging among individuals. We identified 320 upregulated and 90 downregulated genes in aged ovaries. Gene enrichment analyses using ORA and GSEA revealed upregulation of genes associated with oxidative stress, immune system processes, cell-to-cell interactions, and several signaling pathways, which have been reported as drivers of aging in other organs.

In addition, we identified alterations in components of the steroidogenic machinery, such as the well-known ovarian aromatase (cyp19a1a). Reactome and KEGG analyses further revealed enrichment of apoptosis-related and cell signaling pathways. Because ovarian aging is a multifactorial process, we are currently validating several candidate mechanisms using in situ hybridization and immunofluorescence to understand the histological, cellular, and molecular bases of ovarian aging and determining whether all zebrafish ovaries age in the same way

17.12 Nadja Groos - Philipps University Marsburg

Title: The role of Tbx20 signaling during the specification of hematopoietic stem and progenitor cells

Hematopoietic stem and progenitor cells (HSPCs) are multipotent cells, which give rise to all blood lineages throughout life. During the definitive wave of hematopoiesis, HSPCs arise from a specialized subset of endothelial cells (ECs) in the ventral wall of the dorsal aorta (VDA), known as hemogenic endothelial cells (HECs), through a process called endothelial-to-hematopoietic transition (EHT). While the transcription factors Gata2 and Runx1 have been well-established as key regulators in the development of definitive HSPCs, the mechanisms that restrict endothelial cell fate decisions remain poorly understood. We hypothesize that the transcription factor Tbx20 is involved in balancing vascular and hematopoietic identities in the DA, downstream of Apelin signaling. Recent publications have revealed that only aplnr-non-expressing ECs serve as hemogenic endothelial precursors and give rise to HSPCs. Loss of Apelin signaling leads to decreased tbx20 expression and, furthermore, the loss of either Tbx20 or Apelin signaling results in increased HSPC numbers. Findings so far suggest, that Apelin signaling regulates HSPC formation through Tbx20 by preserving endothelial identity. By identifying regulators of HSPC transition, this project will provide fundamental insights into hematopoietic stem cell development and enhance our understanding of the molecular mechanisms that govern cell fate decisions during development and regeneration.

17.13 David Bierbach - Humbolt University zu Berlin

Title: The naturally clonal Amazon Molly (Poecilia formosa) as a model species in ecology and evolution

The Amazon molly (Poecilia formosa) - named after the female warrior tribe in greek mythology - is a naturally clonal vertebrate that has become an exceptional model system for addressing fundamental questions in ecology, evolution, and animal behavior. Originating through an ancient hybridization between two closely related sexual species, this all-female fish reproduces by Gynogenesis, producing genetically near-identical offspring while requiring sperm from closely related species to initiate embryonic development. This unique reproductive strategy provides a rare opportunity to disentangle the effects of genotype and environment on phenotypic variation, enabling experimental approaches that are often impossible in genetically diverse populations. In this talk, I will present how using the Amazon molly as a model has advanced our understanding of fundamental ecological and evolutionary processes. By exploiting the genetic uniformity of clonal lineages, our work reveals how individual differences in behavior, cognition, physiology, and social interactions emerge despite minimal genetic variation. This allows us to quantify the contributions of developmental processes, environmental conditions, and social feedbacks to the generation and maintenance of consistent individual differences and collective dynamics. Combining controlled laboratory experiments, high-resolution behavioral tracking, genomic tools, and computational modeling has enabled us to explore mechanisms operating across biological scales—from individual differences to population-level patterns.

17.14 Masaru Matsuda - Utsunomiya University Japan

Title: Improvement of the recipient strain for germ cell transplantation in medaka cryopreservation

In fish, although sperm can be cryopreserved, this method is not applicable to eggs and fertilized eggs. In the context of preserving genetic resources from inbred lines or wild populations that necessitate both males and females, it is imperative to maintain these entities in a living state. Conversely, in the transplantation process, germ cells extracted from the ovaries or testes are introduced into sterile hatching fry, which lack the capacity to produce their own germ cells. As a result of this procedure, the transplanted germ cells are integrated into the gonads of the hatching fry. Given the absence of germ cells in these larval fish, the production of offspring is constrained to sperm and eggs derived from transplanted germ cells. Despite documented attempts at germ cell transplantation in medaka, with reported successful cases, there remains a need for further refinement in establishing a recipient line that lacks the capacity to produce germ cells autonomously. In this study, we report the successful development of a medaka recipient line that is optimal for germ cell transplantation. To obtain infertile fry, a cross was designed between individuals heterozygous for the dnd gene mutation. It was expected that 25% of the offspring would be homozygous for the mutation, resulting in the absence of germ cells and infertility. However, it was impossible to determine which of the fry were infertile. Consequently, the dnd mutation was introduced into the genetic background of a transgenic line that expressed green fluorescent protein (GFP) in its germ cells. This facilitated the identification of infertile fry, which were characterized by the absence of GFP expression in the gonadal region of the hatched fry. However, when germ cells were transplanted into this line, all individuals differentiated into males, irrespective of their genetic sex. Given the documented finding that the number of germ cells during early development exerts a significant influence on an individual's sex in medaka, a hypothesis was formulated proposing that differentiation into males occurred due to an inadequate number of transplanted germ cells.

Consequently, a mutation was introduced into the anti-Müllerian hormone receptor II (amhrII) gene of this strain, which has been demonstrated to increase the number of germ cells and facilitate sex reversal (feminization) in genetically male individuals. Consequently, when germ cells were transplanted into dnd homozygous and amhrII homozygous fry, these individuals differentiated into males or females, contingent on the sex of the fry, and produced donor-derived sperm or eggs. The efficacy of this strain as a recipient in germ cell transplantation was demonstrated, leading to the differentiation of germ cell-transplanted fry into males or females, depending on their genetic makeup. The cryopreservation of testes from inbred and wild strains, followed by the transplantation of cells dissociated after thawing into the hatched fry of the recipient strain, has enabled the revival of individuals derived from the donor germ cells. This facilitates crisis management in the face of the loss of living resources and the revival of specific strains in regions where transporting live fish is challenging.

17.15 Elise Pesce - Vrije University Amsterdam

Title: Neural and glial cells development in Zebrafish Embryos as an Endpoint of Thyroid-related Developmental Neurotoxicity

Thyroid Hormone System Disrupting Chemicals (THSDCs) are known to affect the development of the nervous system in vertebrates by interfering with the thyroid hormone system (THS) during early-life development. THs play a crucial role in brain development, including the differentiation of neural and glial cells, and myelinization of oligodendrocytes. This could ultimately lead to impaired cognitive function. As a result, assessing THS disruption effects on brain development is a significant concern for environmental and human health safety.

We are currently investigating the effects of THS disruption using a triple-transgenic zebrafish (Danio rerio) line which allows for the quantification of three different brain cell types and thereby the detection of developmental neurotoxicity (DNT). In this line, neurons, astrocytes and oligodendrocytes express three distinct fluorescent proteins, driven by the regulatory sequences of enolase 2, glial fibrillary acidic protein, and myelin basic protein, respectively. Previous studies have shown alterations in oligodendrocytes differentiation following exposure to several DNT compounds and THSDCs. Consequently, we are investigating exposure to different THS disruption mechanisms of action, that likely result in structural alterations of oligodendrocytes and neurons development.

After 5-days of exposure to model compounds, T3 (natural thyroid hormone) and Propylthiouracil (TH synthesis inhibitor), we firstly investigated oligodendrocytes structural alterations in zebrafish embryos by confocal imaging. Results show that the volume of oligodendrocytes in the brain decreased in PTU-exposed fish and increased in T3-exposed fish. Secondly, to confirm dysregulation in key genes expression involved in neurodevelopmental processes, we are investigating thyroid- and brain-related gene expression and DNT biomarkers. After validation of the methods using the model compounds, environmental relevant pollutants, known for their THS disrupting properties, will be tested in this model.

These new brain-specific approaches will be combined with specific TH-related endpoints, such as TH levels and thyroid follicle morphology to assess sensitivity and specificity of oligodendrocytes and neurons development as a thyroid-related DNT endpoint.
In summary, our data suggest that new endpoints focusing on DNT could be a valuable addition for THS disruption assessment in zebrafish embryos, as this is done in the recently developed tFET (thyroid Fish Embryo Toxicity test) currently under OECD validation for the assessment of environmental endocrine disruptors.

17.16 Luis Hernandez Huertas - EMBL Heidelberg

Title: Understanding cell-specific proteostasis under temperature stress conditions in zebrafish embryos

Embryonic development is a highly coordinated process influenced by environmental factors such as temperature, oxygen availability, and metabolic state. Understanding how organisms preserve developmental stability under fluctuating conditions remains a fundamental challenge in developmental and systems biology. Recent whole-animal single-cell RNA-seq studies suggest that cellular sensitivity to temperature-induced stress varies across cell types, potentially due to differences in protein homeostasis (proteostasis), which encompasses protein synthesis, folding, and degradation. The primary goal of this project is to investigate the role of proteostasis in modulating cell-type-specific sensitivity and resilience to thermal stress during zebrafish development. We hypothesis that differential cellular responses to stress may be driven by variations in translational dynamics, codon optimality, tRNA availability, and protein stability. To address this, we are performing polysome profiling and single-cell ribosome profiling in zebrafish embryos under both control and temperature-stress conditions, integrated with a proteomic labelling approach to decipher translational regulation. We will also assess codon usage and codon optimality across distinct cell types to determine their relationship with translational efficiency. We anticipate identifying molecular mechanisms that confer either robustness or vulnerability to environmental stress across cell populations. This work will advance our understanding of how proteostasis contributes to stress adaptation, providing insights into how post-transcriptional regulation shape cellular stress responses during embryogenesis.

17.17 Ilayda Piskin - KU Leuven Belgium

Title: Characterizing the age-related neuromuscular decline in short-lived African Turquoise Killifish

Aging drives progressive decline in muscular and neural systems, resulting in reduced physical function. This deterioration underlies sarcopenia, the age-related loss of muscle mass and function, which is a leading cause of frailty and disability in the elderly. Therefore, discovering therapeutic targets is essential. In our study, we employed the shortest-lived vertebrate laboratory species African turquoise killifish as a gerontology model to characterize neuromuscular aging.

We examined age-related changes in three components of the neuromuscular system: skeletal muscle, the spinal cord, and the neuromuscular junction. Using behavioral, molecular, and bioelectrical approaches, we investigated these changes at multiple levels. Through swim endurance and respirometry assays, we demonstrate that motor function and metabolic rate decline with age. We further described age-related alterations in muscle composition and membrane integrity using electrical impedance myography. Finally, we assessed neuromuscular junction integrity and motor neuron health, revealing signs of denervation.

Together, our findings establish that killifish undergo neuromuscular deterioration, supporting its utility as a preclinical model for developing disease-modifying therapies for age-related diseases.

17.18 Carmen Feijoo - Andres Bello University, Chile

Title: Early intestinal macrophage populations shape the establishment of oral tolerance to dietary antigens

Food allergy results from a failure to establish tolerance to dietary antigens during early life. Oral tolerance is an active immunological process through which innocuous food antigens are recognized and accepted rather than eliciting inflammatory immune responses. This process takes place primarily in the small intestine, where dietary antigens first encounter epithelial and immune cells within a highly specialized tissue environment. The weaning period, when solid food is introduced and the intestinal immune system undergoes rapid maturation, represents a critical developmental window for the establishment of long-term tolerance to dietary antigens. Clinical and experimental evidence indicates that recurrent infections and inflammatory episodes during this period can disrupt immune maturation and increase susceptibility to allergic diseases, including food allergy. However, the mechanisms that enable the intestine to identify food antigens as non-threatening remain not fully understood.

Using zebrafish as an in vivo model, we investigated the contribution of intestinal macrophage (MP) populations to oral tolerance establishment and the impact of early-life inflammation on this process. Single-cell RNA sequencing revealed the sequential appearance of distinct MP populations following the introduction of solid food. Large stellate f13a1b+ homeostatic and mfge8+tissue-remodeling MP were the first populations detected in the intestine. These populations were subsequently followed by the emergence of a small mhc-II+ MP population capable of actively taking up and processing dietary antigens. To determine how inflammatory conditions affect this developmental program, larvae were exposed to a pro-inflammatory environment during the period of intestinal MP colonization. Inflammation altered the function of the mfge8+ tissue-remodeling MP and significantly reduced the proportion of mhc-II+ MP capable of taking up dietary antigens. This defect was associated with a reduction in systemic tolerance to dietary antigens.

Ongoing studies are investigating whether the inflammatory environment induces metabolic reprogramming of specific macrophage populations, contributing to the observed defects in antigen uptake and tolerance induction. These results identify specific intestinal macrophage populations as a critical component of early-life immune education and provide new insight into how inflammatory events during weaning may increase susceptibility to food allergy.

17.19 Océane Vanonckelen - KU Leuven, Belgium

Title: The short-lived killifish as a model to study age-related changes in brain advanced glycation end products and proteostasis

Aging is associated with physical and cognitive decline that impairs quality of life, yet the underlying mechanisms remain incompletely understood. Appropriate animal models are therefore essential to capture the complex processes underlying brain aging. We therefore use the short-lived GRZ strain of the African turquoise killifish, an ideal model due to its short lifespan and conserved hallmarks of human aging. However, some key aspects of brain aging remain unexplored in this strain. To further establish its relevance, we characterized age-related accumulation of advanced glycation endproducts (AGEs), alterations in the ubiquitin proteasome system (UPS) and inflammatory markers in the killifish brain. AGEs arise from reactive dicarbonyl species such as methylglyoxal and glyoxal, and they are cleared in part by the UPS, which can switch from the constitutive subunits that constitute the standard proteasome to the inducible immunoproteasome subunits under stress or inflammatory conditions.

We show that AGEs and their precursors accumulate in the aging killifish brain and this is accompanied by an upregulation of inflammatory markers, alterations in the UPS, and reduced proteasome activity. Interestingly, immunoproteasome subunits were expressed specifically in microglia-like cells and were upregulated with aging. To further unveil the regulation of the UPS in the killifish brain, we i.p. injected fish with LPS and quantified expression levels of the different constitutive and inducible proteasome subunits. Contrary to young fish in which all inducible UPS subunits are upregulated in response to LPS, aged fish failed to switch from the constitutive to the immunoproteasome subunits.

To conclude, we here provide the first evidence for disturbed protein glycation and UPS activity in the aging (short-lived) killifish brain. As several of our findings resemble features observed in human brain aging and neurodegenerative conditions such as Alzheimer's disease, they further support the killifish as a valuable model to study mechanisms of brain aging and neurodegenerative disease.

17.20 Rie Kusakabe - Kansai University, Japan

Title: Early development of the Pacific saury (Cololabis saira) and implications for comparative developmental biology

Among teleost fishes, the order Beloniformes includes many well-known species, such as the medaka (Oryzias latipes), an important experimental model, and exhibits remarkable morphological diversity. This order also includes the Pacific saury (Cololabis saira), a commercially important fish species in Japan. Despite its ecological and economic importance, many aspects of its life history and development remain poorly understood.

In this study, we established a standard developmental staging series for Pacific saury. Fertilized eggs were obtained through natural spawning under captive conditions and by artificial fertilization. Embryos were cultured in artificial seawater at 16 °C and observed until the larval stage. Under these conditions, cleavage and gastrulation proceeded more slowly than in medaka. In contrast, somitogenesis began relatively early, and numerous somites had already formed by the completion of epiboly. This feature suggests that the developmental timing of Pacific saury differs from that of other model fish species, such as zebrafish and medaka, and may reflect a species-specific developmental strategy. After hatching, larvae immediately initiated feeding and reached adulthood in approximately six months.

We also examined the cellular basis of the characteristic blue body coloration of Pacific saury. Light microscopy revealed that the blue coloration was not visible under transmitted light but became apparent under epi-illumination. Ultrastructural observations using transmission electron microscopy showed that the coloration is produced by iridophores located in the dermis. These cells contained reflective platelets arranged around the nucleus, a structure known to enhance light reflection and generate vivid coloration. Similar iridophores have previously been reported mainly in tropical and subtropical marine fishes, and our observations suggest that comparable cellular mechanisms contribute to the blue coloration of Pacific saury.

Taking advantage of genomic resources established recently, we are currently investigating the expression patterns of developmental regulatory genes of Pacific saury.

This study provides the first detailed description of early development and physiology in this species. Comparative analyses with other teleost fishes, including medaka, are expected to provide new insights into the diversification of developmental processes and the biology of fish species that inhabit changing marine environments.

17.21 Jean Eberlein - Marburg University

Title: Apelin signaling acts as a molecular switch between endothelial and hematopoietic stem cell fates

Hematopoietic stem and progenitor cells (HSPCs) emerge from arterial endothelial cells (ECs) through a process termed endothelial-to-hematopoietic-transition (EHT), a process induced by paracrine signals and driven by a transcriptional cascade. Despite inductive signals being broadly received by ECs in the dorsal aorta (DA), only a subset of ECs undergoes EHT, while others maintain their vascular identity. The molecular mechanisms that determine this selective fate decision remain poorly understood. Here, we discover Apelin signaling as a critical regulator of cell fates in the DA, acting as a molecular switch to balance vascular and hematopoietic identities. We show that Apelin receptor (Aplnr)-expressing ECs retain their arterial identity, while aplnr non-expressing ECs are primed to become hemogenic endothelial cells (HECs) and transition into HSPCs. Loss of Apelin signaling leads to excessive EC-to-HEC conversion and increased HSPC numbers. Conversely, forced Aplnr expression abolishes HSPC formation by maintaining EC identity. These findings reveal that Apelin signaling regulates HSPC formation by preserving endothelial identity. In summary, our findings establish Apelin signaling as a critical regulator for balancing endothelial and hematopoietic fates.

17.22 Fiona Carey - IMB Mainz

Title: Buc2l Drives Higher-Order Germplasm Assembly in Zebrafish via a Conserved Tandem Repeat Region

Germ cell fate in zebrafish is specified by inheritance of germplasm, a maternally deposited, phase-separated compartment first assembled as the Balbiani body during oogenesis and later reorganized at cleavage furrows of the early embryo. Bucky ball (Buc) is the central organizer of this process, but the full complement of factors driving germplasm condensation remains unclear. Here we characterize Bucky ball-2-like (Buc2l), a previously uncharacterized, disordered, RNA-binding paralog of Buc conserved across teleosts. Buc2l lacks Buc's canonical prion-like domain but contains a tandem repeat motif whose copy number varies across species while its position and sequence composition remain conserved, suggesting selection for a repeat-based multivalency module. Buc2l colocalizes with Buc and other germplasm components in oocytes and embryos and interacts with Buc, Ziwi, and Tdrd6a/b. Using CRISPR/Cas9-generated mutants, we show Buc2l is dispensable for Balbiani body formation, oocyte development, and fertility, but is required in the embryo for aggregation of germplasm into large, organized granules without it, Buc and other components remain dispersed and fragmented, maternal buc and dazl mRNAs fail to be retained, primordial germ cell numbers collapse, and mutant fish develop exclusively as sterile males. Biochemically, recombinant Buc and Buc2l each undergo concentration-dependent liquid–liquid phase separation and cooperatively form larger, more stable condensates together than either alone. Deletion analyses show the tandem repeats are necessary and sufficient for Buc2l self-assembly and that condensate size scales with repeat number. Together, these data identify Buc2l as a stage-specific amplifier of germplasm assembly, revealing repeat-driven multivalency as a distinct, conserved mechanism for building functional germ granules and safeguarding germline integrity.

17.23 Snigda Bhardwaj - COS, Heidelberg

Title: Investigating the Mechanisms Underlying Interspecies Chimerism in Teleost

Interspecies transplantation provides a robust experimental system for investigating fundamental principles of developmental biology, including how cells behave within a foreign embryonic environment. However, the generation of successful interspecies chimeras remains challenging due to multiple layers of incompatibility that increase with evolutionary divergence. One potential barrier to successful donor cell integration is incompatibility in cell-cell adhesion, which may impair cell mixing and promote segregation within the host embryo.

Using blastula-stage transplantation across multiple teleost species, we observed reduced donor cell dispersion and increased clustering as the evolutionary distance between donor and host species increased. These segregation patterns suggest that differences in adhesive properties contribute to donor cell exclusion from the host embryonic environment. To investigate this possibility, we focused on cadherin-mediated adhesion, a key regulator of cell-cell interactions during early development. Experimental modulation of E-cadherin expression in donor cells was associated with enhanced integration and improved dispersal within host tissues, supporting a functional role for cadherin-mediated adhesion in overcoming species-specific incompatibilities. Together, these findings support the hypothesis that cadherin-mediated adhesion contributes to the species-specific regulation of cell mixing during early morphogenesis and provides a framework for further investigating adhesion-based mechanisms underlying interspecies compatibility.

17.24 Marie Pardon - KU Leuven, Belgium

Title: The Zebrafish Model as a New Approach Methodology (NAM) for Multi Toxicity Assessment of Aromatic bio-based Alternatives to Substances of Concern

The extensive use and environmental release of substances of very high concern (SVHCs) resulting from global industrialization pose significant risks to both human health and ecosystems. Aromatic SVHCs such as bisphenol A (BPA), nonylphenols, and alkylbenzene sulfonates are widely used in plastics, can coatings, surfactants, and flame retardants, leading to continuous exposure across environmental compartments. Developing safer, bio-based alternatives derived from renewable resources, offers a promising strategy to reduce the adverse impacts associated with these compounds. Aquatic vertebrate models are increasingly employed to evaluate chemical safety while simultaneously providing insight into potential ecological effects. The zebrafish (Danio rerio) is particularly valuable for toxicological assessment due to its optical transparency during embryonic development, which enables direct visualization of organogenesis and physiological processes. Moreover, the use of zebrafish embryos up to 120 hours post-fertilization (hpf) constitutes an ethically favorable New Approach Methodology (NAM), bridging the gap between in vitro assays and conventional mammalian testing while supporting high-throughput screening. In this study, the zebrafish model was applied as an integrated platform for multi-toxicity assessment of aromatic SVHCs and their bio-based alternatives. Acute toxicity was evaluated using the zebrafish embryotoxicity test (ZET) to determine EC_{50}, LC_{50}, and the teratogenic index (TI = LC_{50}/EC_{50}). Neurotoxicity was assessed through larval behavioral analysis, focusing on anxiety- and stress-related responses during light–dark transition assays. Our results demonstrate that several aromatic SVHCs exhibit pronounced teratogenic effects and induce neurotoxicity at concentrations below those causing overt morphological abnormalities. Notably, many biobased alternatives displayed a more favorable toxicity profile across the evaluated endpoints, highlighting their potential as safer and more sustainable substitutes. Collectively, these findings support the current classification of these chemicals as SVHCs and further validate the zebrafish model as an effective NAM for comprehensive, multi-endpoint toxicity assessment of chemicals of concern.

17.25 Maria Alexandra Giannopoulou - Tron Translational Oncolgoy Mainz

Title: Modulation and characterisation of Tunneling Nanotubes in vitro and in Zebrafish embryo

Advances in cancer research have improved survival, but therapy resistance remains a major barrier to complete remission and recovery and is a leading cause of death in treated patients. Resistance can arise from tumor cell–intrinsic changes (e.g., numerical or structural alterations of drug targets) and from interactions between tumor cells and the tumor microenvironment (TME). Increasing evidence also implicates intercellular communication in cancer progression and treatment failure (Worzfeld et al. 2017). Beyond classical signaling via secreted factors and direct contact mechanisms such as gap junctions or synapses, cells can form tunneling nanotubes (TNTs), first described in 2004 (Rustom et al. 2004; Lou et al. 2012). These actin-based physical connections enable long-distance transfer of organelles and other intracellular components, potentially enhancing tumor invasiveness, adaptation to therapy, and resistance while reducing immune cell effectiveness (Lou et al. 2012; Rehberg et al. 2016; Osswald et al. 2015; Kretschmer et al. 2019; Valdebenito et al. 2020; Valdebenito et al. 2021; Saha et al. 2022). Our work aims to characterize TNT formation and regulation and to develop tools to manipulate TNT networks in order to define how TNT-like structures contribute to therapy resistance. We focus on TNT-like structures at the blastula stage of the zebrafish embryo, whose transparency enables straightforward imaging allowing comparison of TNT formation mechanisms observed in vitro with those in vivo. To disrupt these structures in zebrafish embryos, we will apply TNT-targeting approaches identified in vitro, test their efficacy in vivo, and assess how intercellular connections influence therapy resistance.

17.26 Clara Kempkens - Vrije University, NL

Title: Endocrine disruption of zebrafish olfactory system development

Thyroid hormones (THs) are crucial for neurodevelopmental processes of vertebrates, making them potential targets of environmental pollutants that act as endocrine disruptors. Accordingly, impaired neurodevelopment is considered the primary adverse health effect of TH system disrupting chemicals (THSDCs) in humans, while it has not been sufficiently addressed from that perspective in wildlife, such as fish. A recently established adverse outcome pathway (AOP) shows the link between lowered TH levels and impaired eye development resulting in impaired vision and reduced survival of fish (AOP #365). However, the impact of THSDCs on other neurodevelopmental processes, such as brain and sensory system development have not sufficiently been studied in fish. Consequently, the present PhD project aims to fill these knowledge gaps, focusing, amongst others, on the olfactory system of zebrafish. The findings are set into an environmental context by investigating the impact of THSDCs on development and function of the olfactory system in developing zebrafish embryos. This is done according to the AOP concept to identify critical sensory organ-related endpoints for THSDCs in fish that are also relevant for the risk assessment of other vertebrates and humans. For this purpose, the project is using transgenic zebrafish (tg:sox10) to analyze changes in development (0-5 dpf) of the olfactory epithelium (OE) via fluorescence microscopy. Preliminary results show that model THSDCs induce alterations in the cellular morphology of the OE, an environmentally relevant initial finding. As a next step, we plan to check for the impact of those morphological changes on the olfactory sense of exposed zebrafish.

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17.27 Elena Nicolay - UFZ Leipzig

Title: Development of a high-content analysis workflow to screen the effect of endocrine disrupting chemicals on macrophages and enteric neurons in transgenic zebrafish embryos

Industrially manufactured chemicals are released into the environment, where their potential adverse effects on living organisms are typically not well understood. One particularly sensitive biological target of chemical pollutants is the endocrine system. Current EU regulations state that Endocrine Disrupting Chemicals (EDCs) must be avoided. However, while there are a range of cell-based testing systems to detect potential EDCs, there are a lack of alternative models that consider the full complexity of the endocrine system. Here, we developed an alternative zebrafish embryo-based assay to detect potential EDC effects on intestinal inflammation and enteric nervous system (ENS) development, both targets of ingested EDCs. Using Tg(mpeg1:mCherry) zebrafish embryos, we developed an imaging and analysis workflow to detect region-specific inflammation based on macrophage count and shape, derived from three dimensional (3D) images. The workflow automates anatomical region detection in transmitted light images using the AI-based nnU-Net model to distinguish structures. We then applied Voronoi-Otsu labelling of cells in regions of interest by combining pre-processing, thresholding, and watershed segmentation algorithms to automatically identify individual macrophages in 3D fluorescence images. To establish the screening assay, multiple reference chemicals and exposure windows were assessed. Exposure to 0.15% Dextran Sodium Sulfate (DSS) from 3-5 days post fertilization (dpf) increased intestinal macrophages by 10%. Bisphenol A was selected as a prototypical EDC. Exposure to 10 uM BPA from 3-5 dpf resulted in morphological changes in macrophages, including increased number, size and volume, which indicates an enhanced proinflammatory state. Intestinal macrophage infiltration supports normal ENS development.

In line with previous work, exposure to 1 µM mycophenolate from 2-3 dpf caused a 35% reduction in ENS cell counts in 5 dpf Tg(phox2bb:GFP) larvae. To adapt this assay for double transgenic Tg(mpeg1:mCherry) X Tg(phox2bb:GFP) embryos, we optimized the exposure conditions to 0.15% DSS and 0.5 µM MPA during the period from 2 to 5 dpf. As part of the HORIZON ENDOMIX project, future work will examine whether exposure to mixtures of co-occurring EDCs identified in human cohorts alters macrophage functionality or ENS development.

17.28 Jess Bourn - EMBL Heidelberg

Title: Single-cell mapping of heterochrony to a species-specific phenotype

Vertebrate embryonic development and its underlying genetic programs are highly conserved, yet the rates at which these programs proceed vary greatly between species. Such differences manifest as heterochrony – changes in the relative timing of developmental events – but the extent of such temporal shifts and their relationship to whole-organism developmental tempo remains understudied. To investigate cell-type-specific developmental timing at the molecular and cellular level, we use individual embryo sci-RNA-seq to compare zebrafish and medaka, two poikilothermic vertebrates with a ~3-fold difference in developmental rate. We assemble a time-resolved atlas of medaka from blastula to hatch, comprising >1.1 million cells from >500 embryos and create a cross-species timing resource that places embryos of both species on a shared transcriptional timescale. Using this cross-species framework of development, we identify cell lineages that display notable timing deviations from the predicted whole-embryo alignment between zebrafish and medaka. We define a new, generalisable metric, the “cell-embryo deviation”, for measuring heterochronic shifts in cell type emergence and maturation, and use it to show that many cell types display delay and acceleration that differs from the globally expected slowdown in medaka. Focusing on the notochord, we use in vivo imaging to validate novel timing differences predicted by our framework, and reveal that two cell types which originate from the same progenitor pool exhibit opposing temporal shifts, with one lineage being accelerated and the other delayed relative to zebrafish. Furthermore, we use our single-cell data to identify genes associated with these timing shifts, suggesting candidate mechanisms that may result in notochord-specific deviations from whole-embryo tempo. Together, our findings establish a quantitative framework for predicting and testing heterochrony across species, and suggest that evolutionary changes in developmental timing acts at the level of cell types to remodel tissue morphology and function, thereby acting as a driver of morphological variation.

17.29 Laura Mayer - University Hospital Ulm

Title: Fbxo5 knock-out highly affects early cardiogenesis in zebrafish

Cardiovascular diseases are the leading cause of death worldwide, yet the molecular mechanisms regulating homeostasis and proliferation in cardiomyocytes remain undetermined. Considering this issue, the zebrafish is a powerful model organism for studying cardiac development due to its genetic manipulability and regenerative capacity.
After cryo-injury of adult zebrafish hearts, Klett et al. (2019) discovered an elevation in several cell cycle factors. One of these factors is f-box only protein 5 (fbxo5), suggesting that it plays a role in heart regeneration and potentially also during development. Therefore, this project aims to elucidate the role of fbxo5 in zebrafish heart development.
Fbxo5 mainly regulates the anaphase-promoting complex/cyclosome (APC/C) complex, an E3 ubiquitin ligase. This complex ubiquitinates proteins important in spindle assembly, DNA replication and mitotic exit, representing one of the most important complexes regulating the cell cycle.
Phenotypical analysis of fbxo5 morphants and the fbxo5ti245 mutant line revealed severe malformations of the fish body with heart edema. In morphants the heart is smaller with a reduction in heartbeat rate and ventricular fractional shortening. Whereas homozygous mutants barely show a visible heart and a stronger overall phenotype. The proliferation of cardiomyocytes is significantly reduced in morphants, but no increase in apoptosis is detected. These observations lead to the hypothesis that loss of fbxo5 affects early cardiogenesis. Regarding early heart development, in situ hybridization of cardiomyocyte markers (myl7, cmlc1, vmhc) in fbxo5ti245 mutants reveals defects in heart development in early somite stages. Quantification of nkx2.5 expressing cells shows a decrease in cardiomyocyte progenitors in early developmental stages.
In summary, the given data reveal a severe effect of fbxo5 knock-out on zebrafish heart development in early stages. Further experiments will include a detailed analysis of pathways affected by fbxo5 knock-out in early cardiogenesis and progenitor differentiation.

17.30 Joanna Michowicz - Institute of Molecular Biology, Mainz

Title: Assembling the Balbiani body: Alg13 and its links to translational repressor regulation

The storage and timely degradation of maternal mRNAs are essential for oocyte development, maturation and to support early embryonic development. The Balbiani body (Bb) is a transient oocyte-specific membrane-less organelle rich in maternal mRNAs and RNA-binding proteins as well as membrane-bound organelles. Tight regulation of many RNA-binding proteins is critical to ensure maternal mRNA stability and metabolism. Post-translational modifications, including ubiquitination, can regulate germ granule properties and translational repression activity of residing RNA-binding proteins, but its role in the Bb has not been examined.

We have investigated Alg13, an OTU deubiquitinase that is expressed in zebrafish gonads and interestingly is a component of the Bucky ball (Buc) interactome, a key regulator of Bb formation. Using CRISPR/Cas9, we generated three alg13 mutant alleles. In two of these alleles, alg13 homozygous fish are all fertile male, implying a defect in oocyte maintenance. For the third allele, we still observe a male-biased sex ratio, with the few females displaying oocyte maturation arrest and infertility. In this alg13 allele, the early-stage oocytes have a highly fragmented Bb which we show originates from Bb components not assembling at the nuclear cleft.

We also observe changes in the ubiquitination status of several RNA-binding proteins, some of which are involved in the translational repression of maternal transcripts. Consistent with this, translational repressors are significantly downregulated in alg13 mutants compared to heterozygous siblings. Buc interactors show loss of canonical, annotated Bb components in the absence of Alg13, alongside significant upregulation of many proteins not previously annotated as Bb components. This suggests that the fragmented Bb is aberrant, and likely no longer serves as an effective storage site for maternal transcripts. We hypothesize that Alg13 regulates several ovarian factors by promoting their deubiquitylation to ensure translational repression and we are following up on several candidates.

17.31 Arsenii Dmitriev - Bionomous Switzerland

Title: Automated multi-channel fluorescence screening of zebrafish larvae using a novel benchtop platform (Sortivo™)

Zebrafish larvae are a cornerstone model in developmental biology, genetics, drug discovery and safety pharmacology, among others. Yet manual sorting and screening under a stereomicroscope remains a bottleneck, it is slow, subjective and poorly traceable. Sortivo™ is a new benchtop platform that automates picking, brightfield and multi-channel fluorescence screening (six channels spanning blue to far-red), sorting and single-entity plating of zebrafish larvae (300 µm–2 mm, 2–7 dpf), producing a standardised, image-linked record for every larva.

Two transgenic reporter lines were imaged with Sortivo™: tg(sox1a:gfp), labelling ventral neural progenitors and neurons, and tg(mnx1:gal4;UAS:RFP;cry:GFP), labelling motor neurons (RFP) and the lens (cry:GFP). Larvae were fixed at 5 dpf and imaged in brightfield and fluorescence, lateral and dorsal orientation. Separately, viability was assessed on live 4 dpf wild-type larvae processed through the full Sortivo™ workflow (picking, screening, dispensing), scored for survival and morphology through 120 hpf.

Sortivo™ produced clear, well-resolved brightfield and fluorescence images for both lines, correctly resolving expected expression patterns. Post-process viability was statistically indistinguishable from the anesthesia-only control.

Sortivo™ enables automated, gentle, multi-channel fluorescence screening of transgenic zebrafish larvae without compromising viability, offering zebrafish labs a standardised, traceable alternative to manual stereomicroscope sorting and a scalable route to higher-throughput phenotypic screens.

17.32 Alexander Froschauer - TU Dresden

Title: Conjoined twinning by fission and fusion in Oryzias latipes

Monozygotic twinning has been studied intensively in humans and reported in many animal species. However, no genetic basis or underlying mechanism has yet been discovered for this rare and fascinating phenomenon. A special malformation of monozygotic twins are conjoined twins that are well-known in humans, and that also have been described in fishes, where oviparous development allows the observation of embryos. We observed frequent monozygotic twinning in a medaka strain and postulate the locus MTwin as the causal genetic basis. A female fish exhibited an extraordinarily high rate of (conjoined) twinning and passed this phenotype into the next generation. MTwin is a maternally inherited phenotype that is potentially based on pre-gastrula fission and fusion.

17.33 Christian Schöllhorn - Uniklinik Ulm

Title: Dissecting the role of Smarce1 overexpression in adult zebrafish heart during regeneration

Molecular pathways that regulate organ growth during embryonic development are often reactivated in adulthood to coordinate tissue repair and regeneration after injury. The molecular mechanisms underlying heart growth during development and regeneration after injury remain incompletely understood, despite their considerable relevance to therapeutic heart repair. In our previous studies, we identified Smarce1, a subunit of the SWI/SNF chromatin-remodeling complex, as a regulator of cardiomyocyte (CM) proliferation. Loss of Smarce1 leads to elevated CM proliferation, whereas myocardial Smarce1 overexpression represses CM proliferation in the developing zebrafish heart. To determine how elevated Smarce1 levels affect adult heart regeneration, we examined Tg(myl7:TetOn-smarce1/AcGFP). This doxycycline-inducible system enables CM-specific overexpression of smarce1. Following ventricular cryoinjury, cardiac regeneration was examined at multiple time points using immunofluorescence and Acid Fuchsin Orange G (AFOG) staining to assess myocardial repair and residual wound size. Smarce1 overexpression in CMs impedes cardiac regeneration, as indicated by reduced myocardial repair and increased residual wound area at 28 days post-injury. These findings demonstrate that overexpressed Smarce1 levels impair adult zebrafish heart regeneration.

17.34 Leonhard Kuhlmann - Leibniz Institute on Aging - FLI Jena

Title: The aging brain accumulates pro-inflammatory lipids, which can be mitigated by long-term Licofelone treatment

Brain aging is characterized by an increase in inflammation, which can contribute to the onset of degenerative processes in aging and disease. Lipid derived mediators of inflammation (oxylipins) have been shown to be dysregulated in human plasma of aged individuals as well as Alzheimer's disease patients. However, if aging alters oxylipin levels in the brain, and whether changes in oxylipin homeostasis contribute to brain aging, remains to be elucidated.

To study age-dependent changes in brain oxylipin levels, we utilized the turquoise killifish, as it is the shortest-lived vertebrate model that can be bred in captivity and exhibits spontaneous brain aging phenotypes such as inflammation and neurodegeneration. Targeted metabolomics revealed that killifish brain oxylipin levels change drastically with age, shifting towards a more pro-inflammatory profile. To functionally investigate the role of oxylipins in brain aging, we treated fish over one third of their lifespan with the dual COX/LOX inhibitor Licofelone, which does not only inhibit production of pro-inflammatory oxylipins via the cyclooxygenase pathway, but also prevents shuttling and metabolization of oxylipin precursors through the lipoxygenase pathway. Metabolomic analysis showed a decrease of oxylipins in intestine and brain upon Licofelone treatment, indicating its efficacy. Additionally, proteomic analysis on the brain revealed dampening effects of Licofelone treatment on tRNA-synthesis, protein ubiquitination and mitochondria related proteins. Taken together, our results indicate that aging induces a pro-inflammatory oxylipin profile in the brain, which can be mitigated by long-term Licofelone treatment. Future research should focus on Licofelone induced effects on mitigation of aging beyond oxylipin metabolism.

17.35 Abhishek Patel - Leibniz Institute on Aging - FLI Jena

Title: Quantitative trait locus analysis reveals the genetic architecture underlying changes in cell composition during killifish brain aging

Brain aging is associated with dynamic changes in cellular composition, with distinct cell populations showing different patterns of expansion, decline, and functional remodeling. While these changes might contribute to loss of tissue homeostasis and age-related cognitive decline, the mechanisms behind changes in relative cell-types abundances remain poorly understood.
Here, we aim to investigate the changes in cellular composition in the killifish brain aging and genetic factors underlying these cell-type dynamics. Using a multi-omics approach, we characterized changes in cellular composition in brain tissue of two closely related killifish species Nothobranchius furzeri and Nothobranchius kadleci, which exhibit divergent brain aging phenotypes, with more pronounced aging phenotypes in N. furzeri. Both killifish species exhibited age-related changes in brain cellular composition, consistent with the patterns reported in human and mice models, including increased brain macrophage abundance and reduced oligodendrocyte abundance with age. Despite these shared age-associated changes, we observed pronounced species-specific differences in cellular composition. Aged N. kadleci exhibited higher proportions of oligodendrocytes and foxg1^{+} neurons compared with aged N. furzeri, whereas brain macrophages were more abundant in aged N. furzeri than in N. kadleci. This pattern is consistent with the more pronounced brain aging phenotype observed in N. furzeri.
To uncover the underlying genetics behind these species-specific differences in cellular composition, a quantitative trait locus (QTL) analysis was performed on the F2-cross hybrid population (n = 506 fish), which display a wide variety of cellular compositions despite being of similar ages. QTL mapping identified multiple genetic loci associated with cellular composition in aged brains, including a shared locus on chromosome 4. This shared locus may point to a common genetic mechanism regulating cellular composition during aging. Functional characterization of this locus could provide further insight into the mechanisms underlying brain aging.

17.36 Frank Pfennig - TU Dresden

Title: Nile Tilapia – a species of worldwide aquaculture as a versatile model for molecular reproductive biology

Due to its size as a food fish (Position No. 4 in global aquaculture production) and as a mouth-brooding cichlid, the Nile tilapia is not per se a predestined laboratory model for research. However, extensive studies on reproduction and physiology generated a wealth of knowledge. Moreover, it became a fascinating subject of investigation regarding the formation of social hierarchies and territorial behavior at all levels of the Brain-pituitary-gonad axis (BPG-axis). Our research focused on the level of the gonads and the pituitary, and we generated extensive expression profiles in dominant and subordinate animals, including miRNA patterns in the testes and pituitary glands. To better understand the complex interactions involved in this process, we characterized long-term stable social communities with respect to their hormonal status and generated comprehensive steroid profiles. As a result, we obtained a fish model exhibiting distinctly strong and reduced activity in the BPG-axis to specific social status, suitable for direct comparisons and further studies. Furthermore, metabolic measurements and biochemical analyses were conducted on the sperm and tissues of animals with different social statuses. Our data indicated context-dependent metabolic pathways underlying reproductive investment. Overall, based on our published data over the years, we suggest Nile tilapia as a suitable model to study reproduction in fish.

17.37 Frank Pfennig - TU Dresden

Title: Multi-photon autofluorescence lifetime imaging microscopy (MP-FLIM) for non-invasive and live metabolic characterization of type A spermatogonia in medaka testis

Throughout the life of a male, haploid gametes arise from undifferentiated, unipotent type A spermatogonia (SgA) from testis. They can serve as a source for adult pluripotent stem cells as well. Intra- and interspecific transplantation of SgA are employed in livestock breeding and have importance for research and species conservation. In order to carry out such sophisticated techniques on living cells, SgA need to be reliably and swiftly identified in the donor tissue. For that purpose, we have established stable transgenic medaka reporter lines. Early germ line cells are labeled in these lines by oct4-promotor driven EGFP fluorescence or vasa-promotor driven Histon2B-mCherry fusion protein. Using the oct4::EGFP-line we determined the proportion of SgA in the testis to be 1–3%. This pool of SgA must be regarded as heterogeneous and includes undifferentiated SgA capable of self-renewal, as well as more advanced SgA committed to differentiation and cycling but with unknown potential for self-renewal and stemness. To shed light into that expected heterogeneity of SgA, we applied a phasor MP-FLIM approach to medaka testis. Phasor MP-FLIM is known from non-invasive characterization of several kinds of stem cells. This approach is label-free and uses the difference in autofluorescence lifetimes of free and protein-bound NAD(P)H in a cell, a measurement of the metabolic state of the living cell. A high ratio characterizes the glycolytic state of a cell whereas a low ratio is found in cells with mainly oxidative phosphorylation. A correlation between a high free/bound NAD(P)H ratio and the stem cell potential of cells has been shown. In our measurements 161 cells were classified as SgA. One of them showed a clear glycolytic metabolic signature. All other cells were characterized by an oxidative state. We verified our measurements by targeted chemical induction of glycolytic or oxidative condition at medaka testis cells.

Best Posters Committees

Posters evaluation	Members
Scientific Committee	Nick Foulkes, Tamara Tal, Rima Sciauciunaite, Laetitia Preau, Sven Reischauer, Daniela Panakova