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International Congress on the Biology of Fish 2026

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Prepared by Don MacKinlay, Md Mahamudul Hasan Mredul and Clare Kilgour

Plenary Session

Adaptations and Acclimation to the Environment in Fishes: Integrating Basic and Applied Research.

Brauner, Colin J. (*brauner@zoology.ubc.ca*) Oral Presentation
University of British Columbia

Abstract: Dr. Brauner is a Professor at The University of British Columbia (UBC). He obtained his Ph D from UBC, and then conducted a PDF at the University of Aarhus and the University of Southern Denmark. He then returned to Canada as a Research Associate at McMaster University before becoming an Assistant Professor at San Diego State University, California in 2000. In 2003 he returned to UBC as an Assistant Professor where he has remained. Dr. Brauner investigates environmental adaptations (both mechanistic and evolutionary) in relation to gas-exchange, acid-base balance and ion regulation in fish, integrating responses from the molecular, cellular and organismal level. The ultimate goal is to understand how evolutionary pressures have shaped physiological systems among vertebrates and to determine the degree to which physiological systems can adapt/acclimate to natural and anthropogenic environmental changes. This information is crucial for basic biology and understanding how fish, which represent 50% of vertebrate species, can inhabit almost every aquatic environment on the planet. Best known for his fundamental work on the physiology of fishes, the impacts of his work are far reaching, by having direct applications in understanding the early evolution of vertebrates, fish conservation, aquaculture, aquatic toxicology and fisheries management. He has supervised over 70 B.Sc. research projects, 35 M.Sc. students, 16 Ph.D. students and 26 post-doctoral fellows. Among these, 14 have gone on to become Professors in Canada and around the world, each with their own well-established research programs. Dr. Brauner has been co-editor of the multi-volume treatise “Fish Physiology” since 2000 and has received numerous awards including an honorary doctorate from the Autonomous University of the State of Mexico (2024) for career contributions to the field of aquatic biology and aquaculture. He has conducted research and been a co-instructor for international graduate courses in comparative physiology around the globe.

Plenary Session

From Shark Gills to Coral Reefs: Discovering Mechanisms and Developing Solutions

Tresguerres, Martin (*mtresguerres@ucsd.edu*) Oral Presentation
University of California, San Diego

Abstract: My research has progressed from ion and acid-base regulation in crabs and sharks, to cellular ion transport mechanisms in the gill, to the molecular basis of acid-base sensing. It is particularly meaningful to give this plenary at ICBF, where I first presented many of these mechanistic discoveries. What started as a specialized question in fish physiology ultimately revealed broadly conserved and evolutionarily recurrent mechanisms with

implications extending far beyond fishes. By integrating comparative physiology, cellular and molecular biology, evolutionary biology, and field-based environmental research, we have since explored how ion transport and acid-base sensing mechanisms have been repeatedly co-opted and diversified across evolution to regulate biomineralization, symbiosis, and other foundational biological processes. More recently, this work has expanded into symbiogenesis and the emergence of novel biological complexity with global biogeochemical relevance. Through transdisciplinary collaborations, I have conducted field work across tropical coral reefs, temperate coastal ecosystems, and Antarctica to investigate how these mechanisms shape organismal responses to environmental change, including ocean acidification, coral bleaching, microbiomes, early detection of physiological stress, and restoration strategies, while also uncovering mechanisms potentially relevant for translational medicine. As I write this abstract, these connections appear almost unavoidable. But early on, I had little idea where those questions would lead. Doing fundamental mechanistic research is often slow and nonlinear because we cannot intentionally search for mechanisms we do not yet know exist. However, this is what turns fundamental biology into a foundation: without mechanistic understanding, prediction, restoration, and intervention become a house of cards. That foundation will never be complete, and the research needed to strengthen it never ends. Today, new technologies, computational approaches, and artificial intelligence provide unprecedented opportunities to accelerate mechanistic discovery and inform higher levels of understanding, prediction, and intervention.

Plenary Session

Using Mechanistic Physiology to Inform Salmon Conservation

Eliason, Erika (*erika.eliason@dfo-mpo.gc.ca*) Oral Presentation
Fisheries and Oceans Canada

Abstract: Pacific salmon are an iconic species, sustaining ecological, cultural and economic systems throughout their range. Yet across the North Pacific, salmon face numerous conservation challenges arising from both natural (e.g. landslides) and anthropogenic (e.g. warming water temperatures, fishing) stressors. In this talk, I'll describe how our team uses a mechanistic physiological approach to understand the drivers of several key conservation concerns. Examples include: 1) Why do some salmon successfully migrate to reach their spawning grounds while others fail? 2) Why do adult female salmon experience higher mortality rates than males? 3) Why do salmon die after release from a marine angling encounter? I will show how identifying the physiological mechanisms underlying these challenges has informed (or has the potential to inform) management actions aimed at improving salmon conservation outcomes.

Plenary Session

The Importance of Mechanistic Physiology in Addressing Real-World Problems

Wood, Chris (*woodcm@zoology.ubc.ca*) Oral Presentation
University of British Columbia

Abstract: Over the last decade, I have sensed a drift away from mechanistic physiology as the emphasis has moved more to descriptive physiology to address the predicted impacts of climate change, an existential real-world problem. For example, laboratory tests of swimming performance, temperature tolerance and hypoxia responses have tended to focus on metrics defining failure (e.g. Ucrit, CTmax, Pcrit), rather than on understanding why failure occurs. In this talk, I will argue that there is an important role for mechanistic physiology in addressing real-world problems, from my own experience where this approach has already resulted, or hopefully will result, in legislative or industrial changes. This is because mechanistic understanding is very convincing to industry scientists, regulators, and legislators. Examples will include research on the physiological mechanisms by which: (i) acid rain kills fish, which led to the amendments in 1991 to the U.S. Clean Air Act, thereby limiting SO2 and NOx emissions; (ii) waterborne metals kill fish, which led to the acceptance of the Biotic Ligand Model (BLM) concept by the U.S. EPA, and the subsequent worldwide adoption of the BLM as a regulatory tool supported by industry and governments; (iii) increasing salinity impacted the physiology of fish in the largest lake in China, which led to dramatic changes in way that the government manages freshwater supply to the lake; (iv) road salt kills baby salmon, which may result in improved salting strategies; (iv) current practices induce nephrocalcinosis (kidney stones), a devasting welfare and economic issue in salmonid aquaculture, and how this can be prevented.

STRESS I Stress in fish: A tribute to George Iwama

Stress in fish: A tribute to George Iwama

Matt Vijayan and Paige Ackerman

Abstract: George's contributions to the field of stress biology spans from his early research on acid base dysregulation during stress, to more applied work related to aquaculture, including efficacy of different anesthetics, relationships between salinity, metabolism and ion regulation, modulation of the immune system, and the development of biomarkers of stress in aquaculture. His research group was also the first to establish the linkage between the organismal and cellular stress response by demonstrating interactions between cortisol and the heat shock proteins in fish. George contributed to shaping the scientific landscape not just in Canada but also internationally, and held several senior executive positions at various institutions, including Director General at NRC, Canada, the President of the University of Northern British Columbia, and the Vice President of the Okinawan Institute of Science and Technology, to name a few. His enthusiasm for science and the field of stress is reflected in his trainees and collaborators, who continue to move the field forward. This symposium will highlight some of the ways in which George influenced the field of stress in fish, sharing both retrospective contributions and recent research developments. The symposium will consist of presentations from a mix of George's trainees and collaborators, as well as researchers influenced by George's ideas.

STRESS I Stress in fish: A tribute to George Iwama

Everything is connected: stress, fish health, and the lab that shaped my career

Ackerman, Paige (*Paige.Ackerman@dfo-mpo.gc.ca*) Oral Presentation
Fisheries and Oceans Canada

Abstract: From my earliest work on stress and immune interactions in salmonids to my current role in managing health of wild-enhancement stocks in British Columbia, a single theme has persisted: stress and disease are not separate problems, they are the same problem viewed from different directions. From 1992-2004, my MSc and PhD research in Dr. George Iwama's lab examined how immunological challenge and infection alter physiological stress responses. Together with work by those within and visiting the lab, these studies showed that infection itself is a potent physiological stressor, that stressor exposure can modulate disease expression without always worsening pathology, and that sublethal disease can erode "realized performance capacity" long before mortality is obvious. Dr. Iwama trained a cohort of students who became colleagues, mentors, and lifelong family, and who carried integrative stress physiology perspectives into aquaculture, enhancement, policy, environmental studies, education, and beyond. The lab's culture, constant questioning, constant curiosity about mechanisms, and persistence on linking stress responses to whole-animal performance, shaped how I approach everything from disease risk management and biosecurity design to hatchery operating procedures and training materials for staff in Canada's Salmonid Enhancement Program. I will briefly revisit some

key findings in the context of contemporary fish health challenges, highlight the academic family that passed through George's lab, and reflect on how that intellectual community continues to influence practical decisions in salmon enhancement and disease management in BC today.

STRESS I Stress in fish: A tribute to George Iwama

Science and socials - a decade of adventures with the Iwama lab

Morgan, John - Oral Presentation
Vancouver Island University

Abstract: This presentation will give an overview of my times in the lab of Dr George Iwama in the Dept of Animal Science at UBC from 1988 to 1997, during which I completed my Masters and PhD focusing on the energetic aspects of ion regulation in several species of fish, as well being a research technician developing simple field methods for monitoring stress in salmonids. I will also provide my perspectives on George's style and approach as a thesis supervisor, and how he built a community within the lab and fostered collaborations outside of it that were of tremendous benefit to his students. Finally, I will touch on how he influenced me in my future career as a professor at a teaching-focused university.

STRESS I Stress in fish: A tribute to George Iwama

It's complicated! Incorporating environmental complexity into multiple stressor experiments

Todgham, Anne (*todgham@ucdavis.edu*) Oral Presentation
University of California, Davis

Abstract: Our capacity to predict the effects of global change on aquatic ecosystems requires that we consider the complexity of current and future environmental conditions. Single stressor experiments commonly fail to accurately capture the tolerance of species to environmental change as organisms live in complex, multivariate environments. In addition to examining how organisms respond to multiple co-occurring stressors, other aspects of environmental change such as timing (e.g. serial exposures, duration) and variability (e.g. predictable vs. stochastic, magnitude) may be important components of environmental complexity structuring an organism's response to change. Furthermore, the capacity of fishes to respond to dynamic changes in their environment changes through ontogeny. This research focus started as a PhD student in George Iwama's lab examining inducible stress tolerance of intertidal fishes and now forms that basis of my research program across many systems focused on understanding and managing stress in conservation and sustainable aquaculture. Our results provide evidence of stressor-induced energetic trade-offs in physiology and behaviour, with insights on mechanisms from cellular energy metabolism, which may identify physiological weak links leading to vulnerability to future ocean change. We lost a dear friend and colleague a year ago. Ros Leggatt and I did our undergraduate degrees together at the University of Guelph and then were PhD students together in

George's lab. I look forward to weaving some of Ros's research and stories into this presentation as I look back on my early years as a fish stress physiologist

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STRESS I Stress in fish: A tribute to George Iwama

Stress in fish or de-stress in me: Lessons learned to identify which genes are most sensitive to chemical pollutants in trout alevins.

Basu, Niladri (*niladri.basu@mcgill.ca*) Oral Presentation
McGill University

Abstract: As a Master's student with Dr. Iwama from 1999 to 2001, I learned three lessons (feasibility, clarity, humility) that have shaped my career, including in the current study. For the past decade I have collaborated with Environment and Climate Change Canada to develop methods to assess chemical pollutants that are efficient, informative, and ethical. We have established a 24-hr test method with rainbow trout alevins that aims to generate data comparable to both fish acute (i.e., 96-hr LC50s) and chronic toxicity tests. To date, 39 chemicals with diverse physical and chemical properties have been tested. A key aspect of this work is dose-response modeling of transcriptomic data to identify a transcriptomic point of departure (tPOD). This tPOD is hypothesized to correspond with apical POD value from chronic fish studies, providing a more efficient, informative, and ethical method. This database is rich and allows us to identify which genes respond earliest, and thus in the spirit of the Iwama lab and its focus on stress responses.Initial investigation of six chemicals identifies five common genes: PFKFB4, LEAP2B, NR1D2A, NT5C3, and TPM1.The most common affected pathways are glutathione metabolism, SNARE interactions, and sulfur metabolism. Analysis of the full dataset is underway.Across this large and complex project, the lessons I learned as a graduate student have been repeatedly reinforced: feasibility in developing a robust and trusted test method; clarity in analysing and interpreting large molecular datasets; and humility when engaging with diverse scientific, regulatory, community, and industry audiences.

STRESS I Stress in fish: A tribute to George Iwama

Is scale cortisol the long-sought biomarker for chronic stress in fishes?

Bernier, Nick[1] (*nbernier@uoguelph.ca*) Oral Presentation
Co-Authors: Williams, Reid[1]; McKinley, Abigayle[1]; Best, Carol[1]; Laberge, Fred[1] and Johnson, Stewart[2]
[1]University of Guelph; [2]Fisheries and Oceans Canada

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Abstract: Scale cortisol is increasingly but perhaps prematurely recognized as a biomarker of chronic stress in fish, since its interpretation and effectiveness across species and stressors remain debated. The dynamics of cortisol accumulation and release in scales, compared with plasma cortisol, remain unclear. We evaluated these dynamics in response to unpredictable chronic stress (UCS) in goldfish, juvenile Chinook salmon, and green sunfish. UCS induced chronic stress in all models and sustained increases in scale cortisol

in goldfish, while it had temporary effects in juvenile Chinook salmon and no effect or reduced levels in green sunfish. Conversely, exposure to combined osmotic and thermal stress led to lasting increases in both plasma and scale cortisol in juvenile Chinook salmon. While juvenile Chinook salmon and green sunfish showed rapid cortisol accumulation in scales after acute stress, the effects were inconsistent across studies in goldfish, and the clearance dynamics varied among species. Our findings indicate that the exchange of cortisol between the circulation and scale matrix is dynamic, and cortisol breakdown and clearance from scales can be influenced by prior stressors. While scale cortisol can reflect plasma cortisol changes and provide insight into recent stress responses in some species, it is not a universal chronic stress biomarker. The scale matrix does not allow for a retrospective historical assessment of stress and is sensitive to quick changes in plasma cortisol, making the impact of capture stress crucial to consider when interpreting scale cortisol levels from wild-caught fish.

STRESS I Stress in fish: A tribute to George Iwama

Transcriptome responses in Atlantic salmon (Salmo salar) and Chinook salmon (Oncorhynchus tshawytscha) livers following microcystin-LR exposure

Ellingson, Brandon[1] (*Brandon.ellingson@noaa.gov*) Oral Presentation
Co-Authors: Greenwold, Matthew[2]; Fast, Mark D.[3]; Speciale, Emily[4] and Shartau, Ryan[5]
[1]NOAA's National Marine Fisheries Service, [2] The University of Texas at Tyler, [3] University of Prince Edward Island, [4] The University of North Carolina at Chapel Hill, [5] Washington State University

Abstract: Harmful algal blooms result in millions of dollars of annual losses for salmonid aquaculture producers in coastal British Columbia, Canada. The predominate toxin, microcystin-LR (MC-LR), is produced by the cyanobacteria Microcystis aeruginosa and is associated with the development of net pen liver disease. MC-LR bioaccumulates in the liver of fishes leading to deleterious effects on growth, immune status, and liver function. Hepatic gene expression was examined in Atlantic salmon and Chinook salmon at several timepoints (6 hours, 24 hours, and 2 weeks) following exposure to MC-LR. Large transcriptome responses were induced by MC-LR in both species (Atlantic salmon; 2362 differentially expressed genes; Chinook salmon; 3194 differentially expressed genes). Genes and pathways associated with MC-LR exposure primarily related to signaling, immune responses, metabolism and apoptosis. These findings complement previous genomic research in other fishes and improve the understanding of the physiological response to MC-LR for salmonids. A panel of candidate genes for each species were identified (Atlantic salmon – 39 genes; Chinook salmon – 61 genes) for future qPCR validation. Differentially expressed genes (n = 19) that overlapped across species were also identified for qPCR validation. These candidate biomarkers have the potential to improve detection of sublethal MC-LR exposure which is associated with harmful algal blooms.

STRESS I Stress in fish: A tribute to George Iwama

Beneficial effects of environmental stress in fish: Integrating stress physiology with aquaculture applications

Nakano, Toshiki (*nakanot@tohoku.ac.jp*) Oral Presentation
Co-Authors: Matsumoto, Keishi; Mun, Kwanghyeon; Haga, Kazumi and Yokoyama, Takehiko
Tohoku University

Abstract: Fish are continuously exposed to diverse environmental stressors, including pollutants, abrupt temperature fluctuations, infectious agents, and husbandry-related challenges. Exposure to such stressors elicits a cascade of biochemical and physiological responses mediated by the neuroendocrine system and cellular stress pathways, including the induction of heat shock proteins. While stress is often viewed as detrimental, its biological consequences depend on the intensity, duration, and context of the stimulus. Our studies in coho salmon (Oncorhynchus kisutch), one of the most valued fish species used in aquaculture, demonstrate that acute heat shock and high dietary doses of the antibiotic oxytetracycline (OTC) induce oxidative stress, reflected by shifts in redox status toward enhanced oxidation. Similarly, dietary inclusion of insect protein, such as black soldier fly (Hermetia illucens) larva meal, altered redox balance and influenced physiological condition in fish. Beyond physical and chemical challenges, even mild handling stress modulated the expression of growth-related genes, highlighting the sensitivity of endocrine and metabolic regulation to environmental stimuli. These findings support the concept that stress responses in fish are not uniformly harmful but can vary along a continuum from distress (negative or bad stress) to eustress (positive or good stress). Controlled exposure to moderate stressors may activate adaptive and hermetic mechanisms that enhance resilience, health, and potentially growth performance. Integrating stress physiology with aquaculture practices, therefore, provides a framework for harnessing beneficial stress to improve the fitness and welfare of farmed fish.

STRESS I Stress in fish: A tribute to George Iwama

Out of Air Again: Effects of Repeated Hypoxic Stress on the Hypoxia Tolerance of Brook Trout (Salvelinus fontinalis)

Stairs, Chandler[1] (*cstairs1@unb.ca*) Oral Presentation
Co-Authors: Sacobie, Charles[1]; Etheridge, Sarah[1] and Gillis, Carole-Anne[2]
[1]University of New Brunswick, [2]Gespe'gewa'gi Institute of Natural Understanding

Abstract: The frequency and severity of aquatic hypoxia are increasing as anthropogenic pressures on aquatic systems intensify. While most fish are likely to experience hypoxic conditions at some point during their life cycle, the frequency of hypoxia exposure is also increasing. In this study, we investigated the intraspecific plasticity and repeatability of hypoxia tolerance in a domesticated population of brook trout (Salvelinus fontinalis). Using oxygen tension at loss of equilibrium (PO2-at-LOE) as a measure of hypoxia tolerance, we

tracked individual performance (n = 56) across five weekly hypoxia challenge tests. Following the final challenge, hepatosomatic index (HSI), cardiosomatic index (CSI), ventricle proportion (VP), and interlamellar cell mass (ILCM) were quantified and evaluated as physiological predictors of changes in hypoxia tolerance over time. Hypoxia tolerance improved progressively across repeated exposures, and individual responses became more consistent, indicating physiological acclimation to low-oxygen stress. Relative index models revealed that individuals with higher CSI, VP, and ILCM showed greater improvements in hypoxia tolerance. Together, these results suggest that hypoxia acclimation is associated with coordinated branchial and cardiovascular characteristics linked to reduced metabolic demand and enhanced circulatory capacity under low-oxygen conditions. As hypoxic events become increasingly frequent in aquatic ecosystems, our findings suggest that brook trout possess the capacity to adjust their hypoxia tolerance in response to recurrent low-oxygen conditions.

STRESS I Stress in fish: A tribute to George Iwama

Operationally simple environmental RNA collection using polyester wool enables accurate detection of physiological gene expression rhythms in cultured fish

Takemura, Akihiro[1] (*takemura@cs.u-ryukyu.ac.jp*) Oral Presentation
Co-Authors: Fukunaga, Kodai[1]; Lu, Siqi[1]; Zhu, Yafan[2]; Nakazato, Mayuko[1] and Hasegawa, Yuka[1]
[1] University of the Ryukyus, [2] Tarim University,

Abstract: Environmental RNA (eRNA) has gained increasing attention as a noninvasive molecular indicator capable of capturing real-time physiological information from aquatic organisms due to its rapid degradation dynamics. In this study, we established and validated a practical eRNA collection approach using polyester wool placed within rearing tanks of Malabar grouper (Epinephelus malabaricus). The polyester wool efficiently recovered free cells and mucus-derived materials released into the surrounding water, enabling subsequent RNA extraction and gene expression analysis. RT-PCR successfully amplified target transcripts includingper2,β-actin,ef1α, andrpl8, while all non-RT controls remained negative, confirming that the recovered signal originated from RNA rather than residual genomic DNA. A time-course experiment demonstrated a transient peak in eRNA abundance four hours after fish handling, followed by a marked decline, a pattern consistent with stress-induced mucus secretion during netting procedures. Furthermore, diurnal oscillations inper2expression detected from eRNA closely paralleled the rhythms observed in tissue-derived mRNA, with comparable acrophases identified by Cosinor analysis. Importantly, eRNA remained detectable for at least eight hours under high-temperature conditions (approximately 29 ℃), indicating operational robustness suitable for aquaculture environments. Collectively, these findings demonstrate that polyester-wool–based eRNA sampling provides a reliable, low-cost, and noninvasive method for assessing physiological states in farmed fish. This approach offers strong potential for integration into real-time health monitoring frameworks in modern aquaculture systems.

STRESS I Stress in fish: A tribute to George Iwama

Different mucosae show differential physiological responses associated to specific stressors

Tort, Lluis (*lluis.tort@uab.cat*) Oral Presentation
Co-Authors: Ruiz, Nuria
Universitat Autonoma de Barcelona

Abstract: Main mucosal tissues, (i.e gills, skin, and gut) constitute an interface between the environment and the organism so they play a central role in the early detection of stressors and in orchestrating endocrine, physiological and immunological modulation. Different types of stressors, such as salinity, nanoplastics, anoxia and crowding, and the duration and / or the intensity of them can produce different stress response outcomes. For this study, the selected species was the gilthead seabream (Sparus aurata) that was subjected to two chronic stressors: salinity, nanoplastics (NPs), and two acute stressors, acute crowding and anoxia. The different stressors tested produced distinct levels of upregulation of glucocorticoid receptors, demonstrating that the endocrine sensitivity of fish varies depending on the nature and intensity of the challenge. Similarly, the immune response varied across treatments, with each stressor eliciting a characteristic pattern of mucosal activation. Importantly, results showed that each mucosal tissue exhibits a specific response profile, indicating a tissue-dependent modulation of both endocrine and immune pathways. The intensity of these responses was closely related to the type of stressor applied, highlighting the importance of mucosa differences and considering stressor-specific impacts when assessing fish welfare.

STRESS I Stress in fish: A tribute to George Iwama

The ameliorating effect of calcium on alkaline stress in fish. Claudin 14 takes the stage.

Wilson, Jonathan[1] (*jmwilson@wlu.ca*) Oral Presentation
Co-Authors: Ferreira, Patricia[2]; Montoya, Xena[1] and Soudi, Najiba[1]
[1] Wilfrid Laurier University; [2] University of Ottawa

Abstract: Exposure to soft-alkaline water (pH 10) imposes osmoregulatory challenges to fishes, as ions (e.g. Na+) are lost resulting in ionoregulatory failure and ultimately death. However, previous studies have shown that increasing water hardness (i.e., [Ca2+]) increases the survival and fitness of fishes transferred to high pH water; although, the mechanisms behind this relationship remain poorly understood. The paracellular movement of ions is tightly regulated by tight-junction proteins such as claudins. In this study, we explored the potential role of Claudin-14 (Cldn14) that has been shown to be regulated by Ca2+ levels and to be a potent blocker of Na+ permeation. We hypothesize that elevated [Ca2+] in the water lead to an upregulation of Cldn14 that reduce the paracellular pathways for the movement of divalent cations and Na+, ameliorating the branchial Na+ loss seen in high pH waters. Here, we report Cldn14 localization in the gills of the Mexican tetra Astyanax mexicanus and its regulation in response to environmental pH and Ca2+ levels. We

found an increase in cldn14 mRNA and protein in the gills of A. mexicanus exposed to hard alkaline water ([Ca2+] 1.25mM, pH10), compared to animals held in soft-alkaline water ([Ca2+] 0.02mM, pH8). These results suggest that Cldn14 may provide a possible mechanistic link between the protective effect of water hardness on the high pH ion imbalance observed in fishes. The arc of this research story started in the early 90s as an undergraduate in George's lab and was rekindled 30 years later. So it goes.

STRESS I Stress in fish: A tribute to George Iwama

Reminiscing George Iwama's influence: Extracellular heat shock protein 70 modulate stress signalling in rainbow trout

Vijayan, Mathilakath M.[1] (*mmvijaya@ucalgary.ca*) Oral Presentation
Co-Authors: Faught, Erin [1,2]
[1] University of Calgary, [2] Radboud University

Abstract: My first exposure to heat shock protein (HSP) research began as a postdoctoral researcher in Dr. George Iwama's lab. This really set the stage for me to conduct studies linking the organismal and cellular stress response in fish. At Waterloo, we started examining the interaction of cortisol and HSPs in regulating the cellular stress response. This was followed by our work on the extracellular HSPs in fish and how they were modulated by stressors. We characterized the presence of HSPs in extracellular vesicles (exosomes) and how they were modulated by acute stressors and chronic cortisol stimulation. Exosomes carry a payload of proteins and RNA and play a role in inter-tissue communication and cellular function. We showed that the intracellular chaperone heat shock protein 70 (Hsp70) is exported into the plasma of rainbow trout (Oncorhynchus mykiss) in exosomes. To test the role of plasma exosomes bound to Hsp70 in cell-to-cell communication, we tagged exosomes with a fluorescent lipophilic dye (DiD) and monitored their uptake in trout hepatocytes in primary culture. Exosomes were taken up by hepatocytes over a 24 h period, and this resulted in the phosphorylation of p42/44 MAPK, but not p38 MAPK or cAMP response element-binding protein (CREB). Interestingly, when these extracellular vesicles were incubated with anti-Hsp70 antibody, the exosome-induced increase in p42/p44 MAPK phosphorylation was abolished. This suggests that Hsp70 may be a key player in exosome-mediated cell-to-cell signalling in rainbow trout. Overall, these findings underscore a role for extracellular Hsp70 in cross-tissue communication during stress in fish.

STRESS I Stress in fish: A tribute to George Iwama

The physiological effects of metals: finding peace in the study of stress.

McGeer, Jim (*jmcgeer@wlu.ca*) Oral Presentation
Wilfrid Laurier University

Abstract: Ion dysregulation is a common response that freshwater fish have when exposed to inorganic forms of many metals. Loss of essential ions, through interference with gill uptake mechanisms has been identified as the cause of acute mortality and this

understanding has been exploited, in combination with geochemical speciation, to develop models for predicting the short-term impacts of metals such as Cu, Zn, Ni, Ag, Cd, Pb and others. Significantly, this research has been developed and applied to improve environmental protection and risk assessments. A number of jurisdictions now have water quality criteria and guidelines based on the contributions of fish physiologists. However, one of the many things I learned from George was that dead fish are not very interesting. He instilled in me the interest in understanding the physiological disruption imposed by environmental stressors and the dynamic responses as fish reestablishing homeostatic control over their physiology. In this presentation I will explore some examples of chronic sublethal responses of fish to metals and how they cope with the ongoing stress. In recent projects we have extended investigations to the rare earth elements, critically essential to our transition to a green economy, but poorly understood in terms of potential environmental impacts. Results suggest that in spite of unique geochemistry compared to many other metals, the rare earths induce physiological effects and acclimation responses that appear to be relatively similar.

STRESS I Stress in fish: A tribute to George Iwama

Influences of embryonic ammonia exposure on Hypothalamic-Pituitary-Interrenal (HPI) axis gene expression in Atlantic salmon (Salmo salar L.)

Alipio, Hanna Ross (*hanna.r.alipio@uit.no*) Poster Presentation
Co-Authors: Tveiten, Helge and Sæther, Bjørn-Steinar
UiT The Arctic University of Norway

Abstract: Environmental perturbation during embryonic and larval stages may influence development and functionality of the hypothalamic–pituitary–interrenal (HPI) axis in teleosts. We investigated whether ammonia exposure during specific developmental windows alters expression of key HPI-axis genes in developing Atlantic salmon (Salmo salar). Embryos were exposed to ammonia (4.14 ± 2.61 mg/L NH4+) during three developmental windows: 20% epiboly (70 day degrees, dd) to eyed stage (300dd) (EPI–EYED), eyed stage to pre-start feeding (900dd) (EYED–PSF), or continuously from epiboly to pre-start feeding. Early exposure (EPI–EYED) resulted in earlier hatching, whereas prolonged exposure (EPI–PSF) delayed hatching relative to controls, indicating stage-specific effects on development. Expression of central (crf1a, crf1b), pituitary (pomc1a, pomc1b), and corticosteroid-associated genes (mc2r, mr, star) were quantified at 70dd, 525dd, and 900dd. At 70dd, crf and pomc transcripts were detectable across treatments, while star expression was largely absent. This may suggest that most of the detected transcripts are maternally derived, as activation of the embryonic genome may not yet be fully initiated. At 525dd, early exposure (EPI–EYED) was associated with elevated expression of crf1a, crf1b, mr and mc2r relative to controls, suggesting an increased sensitivity of HPI-axis to ammonia at this stage. By 900dd, the expression of these genes except crf1a declined in EPI–EYED group, while the expression of the other genes remained relatively stable. This s a possible recovery from the earlier ammonia exposure. These

findings indicate that ammonia exposure during embryogenesis interacts with the HPI axis development in a stage-dependent manner, suggesting critical periods in the development of stress regulatory pathways in salmonids.

STRESS I Stress in fish: A tribute to George Iwama

Hypoxia Tolerance of Salmonids within the Restigouche Watershed: The Role of Thermal Refuges

Stairs, Chandler[1] (*cstairs1@unb.ca*) Poster Presentation
Co-Authors: Sacobie, Charles[1] and Gillis, Carole-Anne[2]
[1]University of New Brunswick, [2] Gespe'gewa'gi Institute of Natural Understanding

Abstract: Aquatic ecosystems are increasingly affected by human-induced environmental changes, including climate-driven shifts in water conditions. Rising temperatures reduce oxygen solubility while increasing oxygen demand in ectotherms, exacerbating hypoxia and placing additional stress on fish physiology. However, thermal refuges - cooler, more stable microhabitats - can buffer fish from thermal stress during warm periods. This study examined how habitat type influences the hypoxia tolerance of salmonids, with the goal of understanding whether fish residing in different thermal environments exhibit physiological adaptations to low- oxygen conditions. We conducted hypoxia challenge tests on brook trout (Salvelinus fontinalis) captured from thermal refuge sites and Atlantic salmon (Salmo salar) captured from thermal refuge and mainstem river habitats. Access to thermal refuges improved hypoxia tolerance in both species, with salmon from mainstem habitats showing greater vulnerability than those from thermal refuges. Brook trout exhibited consistent hypoxia tolerance across refuge sites, suggesting a high degree of acclimatization to stable, oxygen-rich environments. These findings highlight the importance of protecting thermal refuges to enhance the resilience of salmonid populations, particularly in the face of climate change and increasing hypoxic stress.

STRESS I Stress in fish: A tribute to George Iwama

Developmental stage dictates the transcriptional response to chemical euthanasia stress in zebrafish embryos and larvae

Tort, Lluis (*lluis.tort@uab.cat*) Poster Presentation
Co-Authors: Ramos, Juan; Khansari, Ali R. and Balasch, Joan Carles
Universitat Autonoma de Barcelona

Abstract: Drugs such as clove oil, tricaine, and lidocaine have been commonly used as anesthetics, and at high doses, as euthanasics for fish. However, their transcriptional effects on early developmental stages, as is often the case with zebrafish Danio rerio, remain poorly understood. In the present study,we assessed the transcriptional response to these three compounds (clove oil, tricaine, and lidocaine), at euthanasic doses across three distinct developmental stages: gastrulation (6 hpf), pharyngula (30 hpf), and larvae (4 dpf). The transcriptional expression of key genes associated with three biological

processes, cell cycle (caspase, shh, jnk), metabolism (sirtuin, ampk, il1β), and stress (crh, gr, hsp70, and c-fos), was evaluated at three different time points. Our data indicates that genes related to the HPI axis, e.g., gr, crh do not appear to be reliable markers during early developmental stages while c-fos shows greater sensitivity and may serve as a more responsive indicator at these stages. The results also indicate that eugenol (clove oil) had a milder impact on gene indicators, whereas tricaine elicited a stronger response, particularly in phase III. All in all, we conclude that anesthetic or euthanasic protocols should be tailored not only to the species, but also to the developmental stage, particularly in model organisms like zebrafish, where intensive research is conducted during early development.

GROWTH Current trends in growth and metabolism of fishes

Current trends in growth and metabolism of fishes

Brian Small, Brian Peterson and Suraj Unniappan

Abstract: An interplay of numerous external and internal factors orchestrates fish growth, feeding, and metabolism. This symposium serves as a platform to highlight recent breakthroughs in our comprehension of somatic growth and energy homeostasis of fishes. Special attention will be directed towards unraveling the metabolic and hormonal processes that regulate growth, while illuminating the ways in which the environment, fish tissues, and associated pathways interact. We extend an invitation to researchers engaged in fundamental and applied investigations into the regulatory aspects of growth and metabolism of fish, with a special emphasis on encouraging active participation from trainee researchers.

GROWTH Current trends in growth and metabolism of fishes

How Marine Three-Spined Stickleback Modulate Metabolic Pathways in Chronic Hypoxia

Negrete Jr., Benjamin (*bnegrete@zoology.ubc.ca*) Oral Presentation
Co-Authors: Richards, Jeffrey
The University of British Columbia

Abstract: Hypoxia, or low oxygen, is a critical stressor globally that inhibits a fish's ability to make ATP to support aerobic metabolism. The marine three-spined stickleback is a coastal fish that encounters bouts of hypoxia, and is a model organism used in evolutionary research. The goal of this project is to investigate the metabolic and genomic responses to chronic hypoxia within a generation (F0), and impacts across generations (F1). This study highlights findings from the first generation (F0). Adult stickleback underwent intermittent respirometry to determine a critical oxygen threshold, Pcrit, which was used to define the hypoxia acclimation conditions. Stickleback were exposed to hypoxia (~40% air saturation) for two weeks, and their metabolic phenotype was characterized. The effects of hypoxia exposure on hypoxia tolerance were assessed by determining the time to loss of equilibrium (tLOE) at 5% air saturation, and the oxygen tension at which fish experience LOE (pLOE).Hypoxia-exposed fish were sampled for pectoral muscle, brain, and liver for biochemical assays (citrate synthase and lactate dehydrogenase), transcriptomics, and epigenetics, and liver and pectoral muscle were sampled for mitochondrial activity. Stickleback in hypoxia showed increased tolerance to hypoxia, as evidenced by higher tLOE at 5% and lower pLOE. Further, liver tissue showed increases in both citrate synthase and lactate dehydrogenase activity. Mitochondrial activity will reveal changes at the cellular level for ATP generation in liver and muscle tissues to contextualize changes in enzyme

activity. These analyses will be repeated in the F1 generation to assess the impacts of hypoxia across generations.

GROWTH Current trends in growth and metabolism of fishes

Growth reduction and aerobic metabolic changes induced by warming and hypoxia in Gulf of St. Lawrence redfish (Sebastes fasciatus)

Guitard, Joëlle[1,2] (*joelle.guitardbio@gmail.com*) Oral Presentation
Co-Authors: Chabot, Denis[1]; Senay, Caroline[1]; Robert, Dominique[2]and Deslauriers, David[2]
[1] Fisheries and Oceans Canada; [2] Université du Québec à Rimouski

Abstract: Rapid warming and hypoxia in the Gulf of St. Lawrence (GSL) are reshaping environmental conditions experienced by redfish (Sebastes fasciatus), a commercially important species whose fishery reopened in 2024 following massive recruitment. Despite high abundance, individuals now occupy habitats characterized by low dissolved oxygen (20–60% saturation) and ongoing warming, yet their physiological limits under these combined stressors remain poorly understood. By conducting two long-term experiment, we quantified how chronic hypoxia (25–100% O_2 saturation; 105 days) and temperature acclimation (2.5–10.0 °C), under current and projected pH scenarios, affect aerobic metabolism (standard metabolic rate, SMR; maximum metabolic rate, MMR; aerobic scope, AS), hypoxia tolerance (O_2crit), food intake, food conversion efficiency (FCE), growth, and condition. Under hypoxia, food intake, FCE, and growth remained stable down to ~45% saturation, declined at 36–44%, and were significantly reduced below ~30%. This threshold corresponded to a linear decline in MMR and AS, while SMR decreased only modestly, indicating reduced aerobic capacity to sustain growth at low oxygen levels. Temperature increased SMR, MMR, and AS, but growth and FCE declined at warmer temperatures, consistent with elevated maintenance costs. Hypoxia tolerance also decreased with warming. Ocean acidification had minimal effects, except for a modest increase in SMR. Together, these findings indicate that redfish can maintain energy balance under moderate stress, but warming and severe oxygen limitation constrain aerobic capacity and reduce growth efficiency. Redfish in the GSL may already be operating near physiological limits, with implications for future productivity and distribution.

GROWTH Current trends in growth and metabolism of fishes

Towards the sustainable production of more robust and resilient European sea bass juveniles in a climate change scenario.

Gutierrez, Joaquin[1] (*jgutierrez@ub.edu*) Oral Presentation
Co-Authors: Montblanch, Manel[1]; García-Pérez, Isabel[1]; Solà, Roger[1]; Bartrés, David[1]; Sánchez-Moya, Albert[1]; Obrador, Hug[1]; Rodríguez, Inmaculada[1]; Fontanillas, Ramon[2]; López, Mariló [3]; Capilla, Encarni[1]; Chillarón, Josep[1]; García de la serrana, Daniel[1] and Blasco, Josefina[1]

[1]Universitat de Barcelona, [2]Skretting, Barcelona,[3]Cooke-España

Abstract: Aquaculture plays a key role in providing healthy food but faces major challenges like sustainability and climate change adaptation. Improving sustainability relies on the reduction of fish-derived ingredients without compromising growth performance or resilience to environmental stressors such astemperature increasing and acidification. We evaluated the effects of fish meal reduction combined with dietary additives and moderate sustained exercise. Cultured myocytes were used to screen bioactive compounds (antioxidants and taurine), which were included into the diet with 10% fish meal and administered for two months to European sea bass fingerlings maintained under sustained or voluntary swimming. The best growth was reached by fish fed taurine (0.9%) combined with moderate swimming, with increased muscle somatic index and reducedmesenteric fat index. The gene expression of the GH/IGF axis and related signaling pathways was significantly modulated by both diet and exercise. A random selection of the experimental fish was transferred to pilot facilities for 7 additional months, fed with a commercial diet under voluntary swimming, and the enhanced growth trajectory was maintained. When exposed to stress challenges, fish with better growth responded similarly to control fish, suggesting no evident impairment in stress resilience. These results highlight the value of early-life nutritional and exercise strategies to promote optimal growth and robustness in European sea bass under aquaculture conditions.

GROWTH Current trends in growth and metabolism of fishes

Routine metabolic rate across early developmental stages of Atlantic salmon (Salmo salar) in relation to survival and growth

Bair, Halli[1] (*halli.bair@maine.edu*) Oral Presentation
Co-Authors: Morefield, Robert[1] and Hamlin, Heather[1]
[1]University of Maine

Abstract: Embryonic survival has emerged as a concern in a selective breeding program for Atlantic salmon at the National Marine Cold Water Aquaculture Center (NMCWAC), where “eye-up” success has declined in recent years. Preliminary observations indicated that mortality peaks coincided with periods of key cardiac morphogenesis and yolk vascularization processes, suggesting that early physiological constrains may contribute to

family level differences. To evaluate whether early life energy homeostasis relates predicts ontogenetic success, we quantified routine metabolic rate (RMR) in 16 half-sib families across three developmental stages: pre eye-up (26-32 days post fertilization, dpf), post eye-up (47-49 dpf), and alevin (62-67 dpf). Oxygen consumption was measured using closed chamber respirometry over 3-hour trials and converted to mass specific RMR (MgO2 g-1 hr-1). Family level metabolic phenotypes were then related to early growth and survival metrics. We hypothesized that families exhibiting higher survival and accelerated early growth would display distinct metabolic phenotypes during these critical developmental windows. Contrary to expectations, RMR did not differ significantly among families at any stage and was not associated with survival. These findings suggest that early survival differences in this breeding population may not be driven by baseline metabolic rate alone. Ongoing morphometric analyses will test whether subtle differences in early energy allocation emerge later in development.

GROWTH Current trends in growth and metabolism of fishes

Warm and well-fed: how ration availability shapes aerobic scope and growth across temperatures in juvenile Chinook salmon

Zillig, Kenneth[1] (*kzillig@umn.edu*) Oral Presentation
Co-Authors: Cooper, Cassidy[2] and Fangue, Nann[2]
[1] University of Minnesota, [2] University of California, Davis

Abstract: Ectotherms in thermally variable environments face metabolic demands that are tightly coupled to food availability, yet the interactive effects of temperature and ration on fish performance remain incompletely understood. This is particularly relevant for Chinook salmon (Oncorhynchus tshawytscha), a species of conservation concern facing increasing thermal stress in warming river systems, particularly those at their southernmost species range boundary. We tested the hypothesis that food limitation would exacerbate the negative effects of elevated temperature on growth and metabolic performance using complementary laboratory and field approaches. In the laboratory, juvenile Chinook salmon were reared under full-factorial combinations of temperature (11, 16, 20°C) and feed ration (35%, 60%, 100%). In a parallel field experiment, fish were caged across sites with naturally varying food availability. Growth was measured throughout each experiment, and metabolic scope was quantified at one timepoint for each study. Both temperature and ration positively influenced growth in lab- and field-reared fish, with ration showing the stronger effect. In the lab, metabolic rates did not differ among ration treatments at 16°C. However, at 20°C, higher rations significantly improved absolute aerobic scope, indicating that nutritional status enables metabolic performance under warm conditions. Contrary to our initial hypothesis, we found no negative interaction between high temperature and low ration; rather, fish capitalized on the growth-promoting effects of warm temperatures when food was abundant. These findings highlight the importance of prey availability in determining whether warming conditions translate into performance gains or deficits for juvenile salmonids under climate change.

GROWTH Current trends in growth and metabolism of fishes

Growth inhibition under high-density and brain transcriptome analysis in rainbow trout

Fujishiro, Kouyou[1] (*gd18024@student.miyazaki-u.ac.jp*) Oral Presentation
Co-Authors: Nishikawa, Sotaro[1]; Yi, Luo[2]; Shimizu, Munetaka[2] and Miyanishi, Hiroshi[1]
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Abstract: High stocking densities negatively affect fish production, even if water deterioration is prevented and the feeding rate is adjusted according to biomass. However, the mechanisms underlying fish growth inhibition under high-density conditions remain unclear. This study aimed to evaluate the growth performance of rainbow trout and investigate changes in gene expression in the brain that processes sensory information under high stocking densities. Juvenile rainbow trout were reared at five different densities (22–180 fish per 18-L tank) for 60 days. Fish in the 180-fish group exhibited significantly lower body weight than those of the 22-fish group on the 28th day, even though environmental factors such as water quality, dissolved oxygen, and feeding rate were the same for both groups. The final fork length and condition factor indicated growth inhibition in the high-density group, which corresponded with decreased plasma IGF-1 levels. Furthermore, brain transcriptomic analyses of the 22- and 180-fish groups were conducted. Gene set enrichment analysis based on the Kyoto Encyclopedia of Genes and Genomes identified significant alterations in five pathways. Among these, steroid biosynthesis and extracellular matrix–receptor interaction pathways were downregulated in the 180-fish group, suggesting the stagnation of myelination in a high-density environment. These findings provide new insights into the physiological and neural mechanisms underlying growth inhibition and brain responses in environments with many individuals of the same fish species.

GROWTH Current trends in growth and metabolism of fishes

Inflammation modulates central regulation of homeostasis in rainbow trout

Soengas, José L[1] (*jsoengas@uvigo.gal*) Oral Presentation
Co-Authors: Portela-Couñago, Laura[1]; Reji, Steffi[2]; Seiliez, Iban[2]; Blanco, Ayelén M.[1] and Conde-Sieira, Marta[1]
[1]Universidade de Vigo, [2]Université de Pau et des Pays de l'Adour

Abstract: The expansion of aquaculture has led to increased use of high-fat diets to improve growth and reduce production costs; however, in mammals, excessive dietary fat induces hypothalamic inflammation that disrupts central regulation of food intake and energy homeostasis. In fish, while peripheral inflammatory responses to high-fat diets are documented, central inflammation and its impact on feeding regulation remain largely unexplored. In this study, we investigated how dietary lipid content and fatty acid composition influence central inflammatory signaling and neuroendocrine control of

feeding and metabolism in rainbow trout (Oncorhynchus mykiss), a teleost species with well-characterized appetite regulation and high aquaculture relevance. Trout were fed diets differing in total fat and vegetable oil composition, and brain expression of inflammatory markers, including cytokines, and NF-κB, as well as key neuropeptides controlling appetite, including npy, agrp, pomc, and cart, was measured alongside metabolic indicators. High-fat diets induced central inflammation, accompanied by altered neuropeptide expression, indicating that inflammation directly modulates central homeostatic mechanisms. The intensity of the response and its effect on neuroendocrine regulation depended on the fatty acid profile, with unsaturated lipids mitigating inflammation and preserving homeostatic balance. These findings provide novel mechanistic evidence linking diet-induced central inflammation to feeding regulation and energy balance in teleosts. Optimizing lipid quality in aquafeeds is therefore essential to promote sustainable aquaculture, enhance fish welfare, reduce production costs, and maintain efficient growth, providing actionable strategies to improve the health, sustainability, and productivity of farmed fish populations

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GROWTH Current trends in growth and metabolism of fishes

Baseline electrogenic characterization of methionine transport and gut barrier function, and responses to specific anti-nutritional factors in Atlantic salmon (Salmo salar) and rainbow trout (Onchorynchus mykiss)

Subramaniam, Marina (*marinas@uidaho.edu*) Oral Presentation
Co-Authors: Small, Brian and Powell, Madison
University of Idaho

Abstract: Aquafeeds have progressed towards alternative protein sources such as those derived from soy, where soybean meal (SBM) represents a high-protein and low-cost source. However, high inclusion levels of SBM in salmonids has resulted in intestinal dysfunction and soybean meal-induced enteritis (SBMIE), mainly attributed to anti-nutritional factors (ANFs). Further investigating the effects of ANFs on intestinal nutrient transport mechanisms can eventually provide mitigation solutions to incorporate higher inclusion levels of soy in fish feed. Here, we report the baseline characterization of methionine transport and transepithelial electrical resistance (TEER)/conductance along the mid-intestine of Atlantic salmon (Salmo salar) and rainbow trout (Oncorhynchus mykiss) using Ussing chambers, and the effects of select isolated ANFs (i.e.: lectin, soya saponin, and β-conglycinin) on methionine absorption. Since SBM is limiting in methionine, it is crucial to investigate methionine absorption and its potential impedance by ANFs in both species. Specifically, the changes in short-circuit current from methionine transport demonstrated similar absorptive kinetics between Atlantic salmon and rainbow trout along both concentration gradients, suggesting similar transport mechanisms may exist in both. Additionally, in comparison to the other ANFs tested, percent inhibition of short-circuit current was only demonstrated with the sequential addition of saponin (10 – 60μM doses) in both species, but no inhibition was detected with TEER/conductance. This suggests that saponin may have a greater effect on nutrient transport, or effects on gut barrier function may require longer exposure periods from the other ANFs. The continuation of this study

includes evaluating changes in total methionine absorption on the liquid-chromatography mass-spectrometry.

GROWTH Current trends in growth and metabolism of fishes

Central activation of Chaperone-Mediated Autophagy Reduces Appetite by Fine-Tuning Hypothalamic Amino Acid Pools: New Insights from Fish

Reji, Steffi[1] (*steffi.reji@inrae.fr*) Oral Presentation
Co-Authors: J. Vélez, Emilio[1]; Blanco, Ayelén M.[2]; Heraud, Cécile[1]; Véron, Vincent[1]; Dias, Karine[1]; Stella, Alexandre[3]; Burlet-Schiltz, Odile[3]; Schnebert, Simon[1]; Roy, Jérôme[1]; Beaumatin, Florian[1]; Cleveland, Beth[4]; Soengas, José L.[2] and Seiliez, Iban[1]
[1]Université de Pau et des Pays de l'Adour, [2]Universidade de Vigo, [3] Université de Toulouse, [4]United States Department of Agriculture

Abstract: Chaperone-Mediated Autophagy (CMA) is a key lysosomal proteolytic pathway essential for cellular homeostasis and metabolism, with dysfunction linked to various human diseases. While extensively studied in humans and mice, CMA was only recently identified in fish, paving the way for novel and evolutionary research perspectives. Here, we demonstrate a role for CMA in regulating feed intake (FI) in rainbow trout (Oncorhynchus mykiss), a major aquaculture species and a widely used model in numerous research fields, including physiology, evolutionary genetics, toxicology, immunology, and nutrition. Specifically, we observed that feed deprivation induces an increase in the CMA activation score – a reliable proxy for CMA activity – in the hypothalamus, a central brain region involved in the regulation of feeding behavior. To probe its functional relevance, we intracerebroventricularly (ICV) injected the CMA activator CA77.1 and found a significant reduction in FI levels, suggesting a regulatory role for CMA in appetite. Further analysis suggested that CMA may regulate FI partly through changes in hypothalamic free amino acid availability, with ribosomal protein degradation potentially contributing to this mechanism. Through this mechanism, CMA may play a critical role in the precise regulation of satiety and represent a promising target for therapeutic strategies aimed at treating metabolic disorders, as well as for nutritional interventions to improve feed efficiency and promote more sustainable growth practices in aquaculture.

GROWTH Current trends in growth and metabolism of fishes

Do Household Plasticizers Affect Taste Receptors in Extra-Oral Tissues of Female Zebrafish?

Naher, Nazmun[1] (*fhm356@usask.ca*) Oral Presentation
Co-Authors: Unniappan, Suraj[1] and Leite, Bianca de Arruda[2]
[1]University of Saskatchewan, [2]University of São Paulo

Abstract: Di-isopentyl phthalate (DiPeP) is a harmful plasticizer used in household items and has adverse effects on feeding and metabolism. Toxic plasticizers, including DiPeP, might affect metabolism. Extra-oral taste receptors (TRs) are emerging players in nutrient sensing

and metabolism. We hypothesized that DiPeP causes adverse effects on metabolism and influences TRs. The main objective of this study was to test whether DiPeP affects metabolism and TR abundance in zebrafish. Methods and Results: Adult female zebrafish (n = 6/group) were exposed to 0 (control), 31.25, 62.50, and 125 µg/L DiPeP for 1 month. DiPeP (125 µg/L) exposure caused significant body weight reduction ($p<0.005$) and modulated appetite-regulating genes. An increase in anorexigenic melanocortin receptor 4 and nucleobindin 2, and a decrease in orexigenic agouti related peptide were found. These gene expression changes suggest a decrease in food intake due to DiPeP exposure. DiPeP increased cellular triglyceride content in zebrafish liver cells. qPCR found TR isoforms: T1R1, T1R2, T1R3, T2R1, T2R4, and T2R5 in the buccal cavity, gut, liver, and ovary. DiPeP (125 µg/L) reduced T1R1 mRNA in the liver, T2R1 mRNA in the buccal cavity, and T2R4 mRNA in the gut and liver ($p<0.001$, 0.023, and 0.002, respectively). Meanwhile, T2R5 mRNA was increased in the ovary ($p<0.001$). Preliminary Conclusions: These results support the hypothesis that DiPeP adversely affects metabolism and TRs. TR modulation possibly contributes to the adverse effects of DiPeP on metabolism. Significance: The multi-system adverse effects of DiPeP identified here highlight the need to regulate harmful plasticizers in household products.

GROWTH Current trends in growth and metabolism of fishes

Crosstalk between the gut-brain axis and the endocannabinoid system in rainbow trout: Implications for appetite regulation

Blanco, Ayelén M.[1] (*amblanco@uvigo.gal*) Oral Presentation
Co-Authors: Paladea-Rojo, Rocío[1]; Fernandez-Maestu, Cristina[1]; Portela-Couñago, Laura[1]; Soengas, José L.[1] and Conde-Sieira, Marta[1]
[1]Universidade de Vigo

Abstract: Feed intake regulation is a complex process involving multiple factors. Among them, the gut-brain axis (GBA) is a bidirectional communication network between the gastrointestinal tract (GIT) and the central nervous system, integrating neural, hormonal, and immune signals to regulate feeding and energy homeostasis. The endocannabinoid system (ECS), composed of endocannabinoids, receptors, and metabolic enzymes, promotes appetite, enhances food reward, and contributes to energy balance. While GBA-ECS interplay is well documented in mammals, little is known about their interaction in fish. This study first assessed the influence of the ECS on the GBA in juvenile rainbow trout (Oncorhynchus mykiss) by administering a general (rimonabant) or peripheral (AM6545) cannabinoid receptor 1 (CNR1) antagonist. Parameters related to appetite regulation, including mRNA and/or protein levels of brain neuropeptides and transcription factors, and mRNA abundance of GIT hormones, were measured at 2 and 6 h post-treatment. Results revealed time- and tissue-dependent modulation of several GBA components in response to ECS blockade. In a second experiment, the effect of short-chain fatty acids (SCFAs: acetate, butyrate, and propionate)—metabolites produced by the gut microbiota—on ECS gene expression was tested in rainbow trout distal intestinal cells. Propionate upregulated mRNAs encoding the Trpv1 receptor after 2h, suggesting stimulation of ECS activity, while

endocannabinoid-synthesizing enzymes showed reduced expression. At 24h, degradation enzyme mRNA levels were altered. Overall, these results suggest a functional interaction between the GBA and the ECS in rainbow trout. Although preliminary, our findings improve understanding of molecular mechanisms regulating appetite and energy homeostasis in fish.

GROWTH Current trends in growth and metabolism of fishes

Chronic high-fat diet intake disrupts gonadal growth anddevelopment in zebrafish

Mustapha, Umar Farouk (*umarfk.mustapha@usask.ca*) Oral Presentation
University of Saskatchewan

Abstract: ***Purpose*:** High-fat diet (HFD) consumption and obesity are closely associated with metabolic dysregulation and impaired reproductive function. However, the cellular mechanisms causing HFD-associated reproductive dysfunction remain incompletely elucidated. In this study, we evaluated how chronic HFD intake affects the reproductive axis of zebrafish. ***Methodology***: Adult male and female zebrafish (n = 8/sex/group) were fed a standard diet (14% fat) or HFD (25% fat) for 2 months, followed by morphometric, immunohistological and qPCR analysis. ***Results***: In males fed HFD, reduced seminiferous tubule scores and decreased number of mature spermatozoa were observed. Similarly, females showed an increase in oocytes at early developmental stages (1 - 3) in the HFD group compared to the control group. Cytochrome P450 family 17 subfamily A member 1 (*cyp17a1*), an important steroidogenic gene, was significantly downregulated in the HFD fish testis, suggesting impaired steroidogenesis. Similarly, oxidative stress markers, kelch-like ECH-associated protein 1a (*keap1a*) and tumor necrosis factor-alpha levels were reduced, whereas superoxide dismutase 2 and interleukin 1 beta (*il1b*) levels were increased in the HFD group. In the ovary, HFD caused a reduction in key regulators of oocyte development and folliculogenesis, including growth differentiation factor 9*,* bone morphogenetic protein 15, germ cell marker DEAD-Box Helicase 4, and stress-response signalling molecules *keap1a* and *il1b*. This suggests alterations in germ cell functions. ***Conclusion***: Metabolic stress caused by high-fat diets disrupts gonadal growth and development in a sex-specific manner. HFD disrupts key cellular processes, leading to reproductive dysfunction. This highlights the importance of dietary nutritional composition on reproductive success.

GROWTH Current trends in growth and metabolism of fishes

Routine metabolic rate across early developmental stages of Atlantic salmon (Salmo salar) in relation to survival and growth

Bair, Halli (*halli.bair@maine.edu*) Poster Presentation
Co-Authors: Morefield, Robert; Hamlin, Heather
University of Maine

Abstract: Embryonic survival has emerged as a concern in a selective breeding program for Atlantic salmon at the National Marine Cold Water Aquaculture Center (NMCWAC), where "eye-up" success has declined in recent years. Preliminary observations indicated that mortality peaks coincided with periods of key cardiac morphogenesis and yolk vascularization processes, suggesting that early physiological constrains may contribute to family level differences. To evaluate whether early life energy homeostasis relates predicts ontogenetic success, we quantified routine metabolic rate (RMR) in 16 half-sib families across three developmental stages: pre eye-up (26-32 days post fertilization, dpf), post eye-up (47-49 dpf), and alevin (62-67 dpf). Oxygen consumption was measured using closed chamber respirometry over 3-hour trials and converted to mass specific RMR (MgO2 g-1 hr-1). Family level metabolic phenotypes were then related to early growth and survival metrics. We hypothesized that families exhibiting higher survival and accelerated early growth would display distinct metabolic phenotypes during these critical developmental windows. Contrary to expectations, RMR did not differ significantly among families at any stage and was not associated with survival. These findings suggest that early survival differences in this breeding population may not be driven by baseline metabolic rate alone. Ongoing morphometric analyses will test whether subtle differences in early energy allocation emerge later in development.

GROWTH Current trends in growth and metabolism of fishes

Thermal constraints on feeding, growth and metabolism in juvenile Chinook Salmon (Oncorhynchus tshawytscha) in Alaska.

Bhagat, Siya (*sbhagat2@alaska.edu*) Poster Presentation
Co-Authors: Snyder, Augustus; Williams, Emily; Gilbert, Matthew; Wagner-Jones, Amelia; Barreriros Scatamburlo, Larissa and Samuel, Will
University of Alaska Fairbanks

Abstract: Chinook salmon (Oncorhynchus tshawytscha) are vital to Alaska's freshwater and marine ecosystems, commercial fisheries, and Indigenous subsistence livelihoods. Increasing frequency and intensity of heat waves pose significant challenges to Chinook populations, including during freshwater early rearing. Thermal conditions experienced during the juvenile life stage can strongly influence energy requirements, feeding, growth, and survival, impacting the abundance and quality of smolts that reach the sea. Despite extensive research on temperature effects in juvenile Chinook salmon from southern latitudes, there remains a knowledge gap regarding the responses of potentially cold adapted northern Alaskan populations to both acute heat events and long-term climate

warming. To address this gap, we conduct controlled laboratory experiments rearing juvenile Chinook across a broad temperature range with varied food availability to reflect natural conditions. We quantify food consumption, growth rates, feed conversion efficiency, and resting and maximum metabolic rates during prolonged warm exposure. These data are being integrated into temperature-performance and bioenergetics models specific to northern chinook. We hypothesize that juvenile Alaska Chinook salmon experience growth and metabolic performance limitations at lower upper temperatures than southern populations due to local thermal adaptation. Likewise, we predict a cold shift in physiological temperature-performance curves, with increased metabolic costs, reduced energetic efficiency, and constrained growth at elevated temperatures, particularly under limited food availability. These results will provide region specific parameters that can support predictive modelling and evidence based fisheries management under current and future climate conditions in Alaska.

GROWTH Current trends in growth and metabolism of fishes

Bridging lab and field: in situ oxygen uptake measurements match laboratory-derived values in Sebastes fasciatus

Guitard, Joëlle[1,2] (*joelle.guitardbio@gmail.com*) Poster Presentation
Co-Authors: Chabot, Denis[1]; Robert, Dominique[2] and Deslauriers, David[2]
[1]Fisheries and Oceans Canada; [2]Université du Québec à Rimouski

Abstract: Under laboratory conditions, measuring oxygen uptake provides a reliable proxy for metabolic rate. However, this require that fish are captured and transported to the laboratory, which may influence their physiology, and laboratory conditions may differ from the wild render the tests less relevant for conservation research. The objective of the present study was to develop a minimally invasive method to estimate metabolic rates in situ. Two custom-built respirometers were deployed at a depth of 30 meters in the St. Lawrence Estuary (Canada) to measure oxygen uptake in wild redfish (Sebastes fasciatus) over a 24-hour period (n=10). To validate the method, in situ routine metabolic rate (RMR) was compared to standard metabolic rate (SMR) measured on the same individuals in the laboratory (n=4) and on laboratory acclimated S. fasciatus (n=11). Despite natural fluctuations in temperature, light, gut fullness and potential biotic stimuli, RMR measured in the wild were consistently similar to laboratory-derived SMR. These findings demonstrate the feasibility of in situ respirometry for estimating oxygen uptake while maintaining ecological relevance.

GROWTH Current trends in growth and metabolism of fishes

Past seawater experience enhances somatic growth in sturgeons

Miyanishi, Hiroshi (*miyanishi@miyazaki-u.ac.jp*) Poster Presentation
University of Miyazaki

Abstract: Accelerating somatic growth and sexual maturation remains a major bottleneck in sturgeon aquaculture, particularly for caviar production, a key specialty of Miyazaki Prefecture, Japan. Some sturgeon species require at least more than five years to reach reproductive maturity, the development of strategies that shorten the production cycle is of considerable economic importance. This study evaluated the effectiveness of the "seawater experience method," in which juveniles are temporarily acclimated to diluted seawater and subsequently returned to freshwater, in Siberian sturgeon (Acipenser baerii) and Russian sturgeon (Acipenser gueldenstaedtii). To determine the optimal treatment protocol, juveniles were transferred from freshwater to various seawater concentrations (30–100%). Based on survival rates and plasma osmolality, the seawater experience was defined as sequential one-week acclimation at 30% and 50% seawater. While Russian sturgeon could not withstand concentrations exceeding 60%, the gradual 30–50% transition achieved nearly 100% survival in both species, with plasma osmolality stabilizing within three days of transfer. Following the return to freshwater, significant growth promotion was observed from the second month onwards. The seawater-experienced groups exhibited a 25% increase in growth for Siberian sturgeon and a 15% increase for Russian sturgeon compared to the freshwater controls. These results demonstrate that past seawater experience effectively accelerates growth, suggesting that physiological changes during acclimation to diluted seawater promote somatic growth, and providing a promising strategy for increasing caviar yield and shortening the time required to reach sexual maturity.

GROWTH Current trends in growth and metabolism of fishes

Profiling proteomic changes in taurine-treated myotubes of European sea bass

Solà, Roger[1] (*rogersola@ub.edu*) Poster Presentation
Co-Authors: Gutierrez, Joaquin[1]; Martin Perez, Miguel[1]; Montblanch, Manel[1]; García-Pérez, Isabel[1]; Vilanova, Mar[2]; Vilaseca, Marta[2]; Chillarón, Josep[1]; García de la serrana, Daniel[1] and Blasco, Josefina[1]
[1]Universitat de Barcelona, [2]Institut de Recerca Biomèdica de Barcelona

Abstract: European sea bass (Dicentrarchus labrax) is one of the most commercially valuable species in the Mediterranean aquaculture. However, the use of highly substituted diets for this species is hampered due to its highly carnivorous habit. The non-protein amino acid taurine has been previously reported as a potential dietary supplement in fish aquaculture, enhancing both growth and antioxidant capacity. Here, to better understand the signalling response triggered by taurine in muscle, proteomic, phosphoproteomic and protein turnover (pulsed-SILAC) analyses were carried out on cultured myotubes from

European sea bass. 2643 proteins were quantified, of which 267 (10.1%) showed significant changes allowing to discriminate between control and taurine (20µM for 24h) treated cells. Downregulated proteins upon taurine treatment were enriched in processes related to cell development, while mostly protein translation processes were upregulated. 7259 phosphosites were identified, of which 1179 (16,2%) displayed significant differences in intensities between the two groups. Several targets of the mTOR signalling pathway showed increased phosphorylation upon taurine treatment, such as AKT, 4EBP1, and EIF4EBP1, as well as transcriptional factors (e.g., EIF3A) involved in protein synthesis, which results a higher protein synthesis capacity and potential increase in proliferation (e.g., PCNA). These findings support a proteinogenic stimulus of taurine in muscle cells by enhancing mTOR signalling and ribosomal activity, thus highlighting its potential as a dietary additive for stimulating muscle growth in European sea bass.

GROWTH Current trends in growth and metabolism of fishes

Artificial light including blue-green wavelengths promote growth of Rachycentron canadum with mRNA upregulation of appetite, growth axis and lipid synthesis

Zhu, Yafan[1] (*xygz48@hotmail.com*) Poster Presentation
Co-Authors: Takemura, Akihiro[2]
[1]Tarim University; [2]University of the Ryukyus

Abstract: Cues from light, including photoperiod, intensity, and wavelength, can all impact physiology in fish. This study investigated the effect of light wavelengths on growth performance in the cobia Rachycentron canadum, a commercially valuable species in tropical waters. Juveniles were exposed to blue (BL, 466 nm peak), green (GL, 518 nm peak), or natural light (NL) for four weeks which resulted in enhanced body weight (BW) and specific growth rate (SGR) in the blue and green treated juveniles. Transcript levels of key visual opsins were more highly upregulated by blue and green light in the diencephalon, compared to the retina, highlighting the importance of extraretinal photoreception in growth regulation. Comparison of transcript levels of various genes regulating appetite or lipogenesis showed a wavelength-dependent effect on growth stimulation in juvenile cobia. Green light influenced transcript levels of the appetite-stimulating peptide, neuropeptide Y (npy), in the brain, and growth hormone receptor(ghr) in the liver. Whereas blue light increased the transcript levels of an anorexigenic peptide, pro-opiomelanocortin (pomc), in the brain, and fatty acid synthase (fas) in the liver. These results align with adaptation to blue-green-dominated marine environment, emphasizing the valuable, non-invasive tool that spectral optimization can be in aquaculture.

CHANGE II Investigating the interrelated responses of climate change, contaminants, and biodiversity loss in fishes

Investigating the interrelated responses of climate change, contaminants, and biodiversity loss in fishes

Tamzin Blewett and Ivan Cadonic

Abstract: Climate change research has been at the forefront of scientific study for the better part of the last few decades. Although much focus has been put on organisms' responses to thermal stress, fishes have to contend with an assault of multiple different stressors including anthropogenic-induced environmental change. Contaminants can interact as well to hinder an animal's ability to respond to other biotic and abiotic stressors (i.e., temperature, dissolved oxygen, salinity, parasites), resulting in reduction in fitness and wellbeing, and further exacerbated by climate variability. These impacts can be seen at the population level for many fish species, resulting in biodiversity loss across the globe. Fishes from sensitive ecosystems (e.g., Arctic or Antarctic) tend to be understudied but are likely to be particularly vulnerable to multiple stressor interactions; therefore, studying their responses to common climate change parameters is vital to reduce biodiversity impacts. This session will focus on both laboratory and field studies that investigate these multiple stressors within a climate change perspective and seek to answer the question of how fishes are able to cope with changing biotic and abiotic parameters. Research presented will span from molecular to whole-animal studies, with focus on studies that incorporate multiple levels of organization or studies that investigate non-model species from vulnerable ecosystems. We look forward to inviting both early career and established researchers to our session to provide a diverse perspective on fish responses to this crisis

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CHANGE II Investigating the interrelated responses of climate, contaminants, & biodiversity loss in fishes

Environmental Variability in Estuaries Modulates PFAS Exposure and Early Life Stage Development in Fishes

Ackerly, Kerri Lynn (*klackerly@utexas.edu*) Oral Presentation
Co-Authors: Roark, Kathleen; DiBona, Elizabeth, Walsh, Grace and Nielsen, Kristin
The University of Texas

Abstract: Estuarine fishes inhabiting coastal systems near urban centres, industrial facilities, and wastewater discharge sites are exposed to multiple environmental stressors that may influence early life stage (ELS) development and survival. ELS fishes are particularly sensitive to environmental change, yet links between contaminant occurrence in estuarine habitats and biological responses in fishes remain limited. Per- and polyfluoroalkyl substances (PFAS) are persistent and mobile contaminants widely detected

in aquatic environments, but potential impacts on estuarine-dependent fishes under environmentally-relevant conditions are poorly understood. We investigated PFAS exposure and associated developmental effects in estuarine fishes from the Texas Gulf Coast. Water and sediment samples were collected downstream of wastewater treatment plant outfalls to characterize spatial and seasonal variability in PFAS occurrence, relative abundance, and partitioning behaviour within habitats used by estuarine fishes. Distinct PFAS signatures among sites and temporal variation in concentrations indicated dynamic exposure conditions within these systems. Environmentally relevant PFAS profiles identified from field sampling informed laboratory exposure studies in ELS sheepshead minnow (Cyprinodon variegatus), red drum (Sciaenops ocellatus), and southern flounder (Paralichthys lethostigma) - three recreationally and ecologically important estuarine-dependent species along the Gulf Coast. Fishes were exposed during ELS development to environmentally relevant PFAS under controlled laboratory conditions. Exposure resulted in morphological changes associated with reduced fitness, including smaller hatch size and altered energetic reserves. These findings connect environmental exposure conditions with organismal responses and demonstrate how contaminant stress within variable estuarine environments can influence ELS development in fishes, potential implications for recruitment and population structure in urbanized coastal ecosystems.

CHANGE II Investigating the interrelated responses of climate, contaminants, & biodiversity loss in fishes

Physiological and molecular impacts of Nickel exposure during thermal acclimation in Rainbow trout (Oncorhynchus mykiss)

Cadonic, Ivan (*cadonic@ualberta.ca*) Oral Presentation
Co-Authors: Griffin, Robert; Davidson-Yee, Sydney; Poulin, Adrienne[1]; Liu, Boyan; Uhrig, R. Glen and Blewett, Tamzin
University of Alberta

Abstract: Aquatic organisms are under increased pressure due to anthropogenic-induced ecosystem changes. Fishes utilize plastic physiological processes to acclimate to changes in habitat temperature; however, concurrently with increased thermal challenges, fishes are having to contend with increasingly contaminated environments. Processes impacted by toxicological assault could impair acclimation capacity, further increasing risk posed by climate change on biota. For example, metals like nickel (Ni), while vital for green technologies, are becoming more prevalent in water bodies due to increased resource extraction and can induce significant osmorespiratory and metabolic consequences for fishes. It is currently unknown how Ni exposure may similarly alter fish acclimation capacity. Therefore, we investigated acclimation dynamics in rainbow trout (Oncorhynchusmykiss) to understand how contaminant exposure could interfere with physiological responses to increased temperature. Trout were acclimated to +5°C (17°C) water for 28 days, while simultaneously exposed to 0 µg/L, 100 µg/L, 250 µg/L, or 500 µg/L of Ni. Critical thermal maximum (CTmax) was assessed on days 0, 1, 7, 14, and 28 to understand how Ni could mitigate acclimation-associated increases in thermal tolerance. On the same days, gill

tissue was sampled for quantitative proteomics to assess how Ni interacts with acclimation-associated molecular responses, in a time- and concentration-dependent manner. Overall, this study provides evidence for how transition to green technologies to attenuate climate change could instead accelerate ecosystem consequences. Understanding how sublethal exposures to pollutants could reduce the capacity for fishes to contend with expected thermal increases is the first step in developing climate change-relevant water quality guidelines.

CHANGE II Investigating the interrelated responses of climate, contaminants, & biodiversity loss in fishes

Beyond Bottom-up Effects: Suspended Sediment Directly Suppress Reef Fish Assemblages and Herbivore Functions

Schakmann, Mathias (*mssorens@hawaii.edu*) Oral Presentation
Co-Authors: Johansen, Jacob; Brijs, Jeroen, Eggertsen, Linda
University of Hawaii HIMB

Abstract: Climate change is intensifying precipitation extremes across the tropics, amplifying sediment runoff to nearshore coral reefs and compounding pressures from land-use change. While sedimentation effects on corals are well documented, quantitative tolerance limits for reef fishes remain poorly established, leaving uncertainty whether fish declines follow habitat degradation or emerge earlier. Here, we examined how reef fish assemblages respond to suspended sediments, quantified as nephelometric turbidity (NTU), across chronic runoff gradients and episodic runoff events on nearshore reefs around O'ahu, Hawai'i. We conducted year-long underwater visual censuses (558 transects; >52,000 individuals; 59 species) alongside co-located measurements of turbidity, temperature, salinity, and concurrent benthic surveys. Generalized Additive Models consistently identified turbidity as the dominant predictor of assemblage structure. Model-derived thresholds revealed >75% loss of total fish abundance and biomass by 5.1 NTU, independent of benthic composition. Herbivores were disproportionately sensitive, reaching >75% loss in abundance and biomass by 3.8 NTU, with functional groups diverging strongly: browsers were virtually absent above ~2 NTU with 75% loss by 1.7 NTU, scrapers declined sharply with 75% loss by 3.5 NTU, and grazers persisted longer but still experienced 75% loss by 4.4 NTU. These fish-function thresholds fall well below published coral stress benchmarks (≥10–30 NTU), indicating that erosion of herbivory, the key top-down control on algal proliferation, can precede benthic degradation. Our findings demonstrate that climate-intensified runoff may initiate reef biodiversity loss through early suppression of fish function, underscoring the need for turbidity-based management targets that protect reef fish communities under changing precipitation regimes.

CHANGE II Investigating the interrelated responses of climate, contaminants, & biodiversity loss in fishes

Glacial retreat as a vector for mercury (Hg) exposure: Evidence for uptake of glacially derived Hg in a coastal food web

Nielsen, Kristin[1] (*kristin.nielsen@austin.utexas.edu*) Oral Presentation
Co-Authors: Barst, Benjamin[2]; Rivera, Tamara[1] and Nelson, Isabelle[1]
[1]University of Texas at Austin; [2] University of Calgary

Abstract: High latitudes are warming rapidly due to polar amplification, accelerating cryosphere melt and glacial retreat. Glaciers act as long-term repositories for contaminants that undergo long-range atmospheric transport - such as mercury (Hg) - which is characterized by a global biogeochemical cycle that is tightly coupled to climatic factors. The heavily glaciated Gulf of Alaska (GoA) region is losing glacial mass faster than other glaciated regions worldwide, and Hg yields in meltwater streams entering the GoA are among the highest ever reported. However, it has not yet been established whether glacially exported inorganic Hg undergoes microbial transformation to methylmercury (MeHg; the highly neurotoxic and bioaccumulative form) and/or whether it is being assimilated into coastal food webs. The propensity of MeHg to biomagnify means that climate-driven increases in glacial Hg export could pose a significant and growing risk to high trophic level species and human consumers.

To address this knowledge gap, we investigated trends in Hg concentrations across multiple trophic levels in biota from glacially influenced waters of Kachemak Bay, Alaska. Sampling spanned a gradient of glacial coverage across multiple watersheds, encompassing high trophic level fish species and lower trophic level prey species and invertebrates. Glacial coverage, watershed characteristics, foraging behavior, and feeding ecology emerged as significant drivers of Hg accumulation. Critically, these findings provide the first evidence that inorganic Hg exported from glacial meltwater may be assimilated into a coastal food web — highlighting glacial retreat as an emerging pathway for contaminant exposure in vulnerable high-latitude ecosystems.

CHANGE II Investigating the interrelated responses of climate, contaminants, & biodiversity loss in fishes

Copper and a common cold: Understanding the immune system modulation of physiological & behavioural impacts of copper exposure in goldsinny wrasse.

Martin, Sidney[1] (*smartin1@ualberta.ca*) Oral Presentation
Co-Authors: Mélançon, Vincent[2]; Vigneault, Juliane[3]; Cadonic, Ivan[1]; Gradito, Maryane[4]; Åsheim, Eirik[5]; Morgan, Rachael[6]; Roche, Dominique[7]; Binning, Sandra[2] and Blewett, Tamzin[1]
[1]University of Alberta; [2]Université de Montréal; [3]Université du Québec à Rimouski; [4]Deakin University; [5]Swedish University of Agricultural Sciences; [6]University of Bergen; [7]Social Sciences and Humanities Research Council of Canada

Abstract: Organisms encounter a myriad of pathogens, often resulting in an upregulation of the immune system to prevent damage associated with infection. This process is energetically costly, with inflammation and stress being common outcomes in infected individuals. In nature, organisms are confronted with pathogens in conjunction with other environmental stressors, including toxicants. Together, these stressors can impact the host organism's physiology, potentially changing the organism's response to contaminants, as well as infections in the environment. To better understand the impact of these combined stressors on a fish's response to copper & infection two experiments were performed. We assess the effect of a copper (Cu) exposure occurring simultaneously with an immune response in goldsinny wrasse (Ctenolabrus rupestris) by utilizing a lipopolysaccharide injection to stimulate the immune system either before (experiment 1) or after (experiment 2) an acute (96-hour) exposure to either a control (0 µg/L), low (125 µg/L), or high (500 µg/L) Cu treatment. Upon completion of each experiment behavioural trials were performed to investigate activity and exploration behaviours. In addition, tissues associated with Cu uptake & accumulation (liver, gill, intestine), as well as those associated with the immune response (spleen) were collected. These tissues were then used to assess the tissue-specific bioaccumulation of Cu and immune gene expression. Our study will help elucidate how physiology and behaviour may be altered when marine organisms are required to withstand multiple stressors.

CHANGE II Investigating the interrelated responses of climate, contaminants, & biodiversity loss in fishes

Will wildfire ash smoke the salmon? Disentangling acid-base and contaminant impacts in UK and Australian fishes

Kwan, Garfield[1] (*gkwan09@gmail.com*) Oral Presentation
Co-Authors: Kirk, Eleanor[1]; Bernat, Tatum[2]; Ibbini, Ziad[3]; Smik, Lucas[1]; Codling, Gary[1], Page, Eleanor[1]; Cramp, Rebecca[4]; Whitehead, Andrew[2]; Fangue, Nann[2]; Franklin, Craig[4]; Tills, Oliver[3]; Wilson, Rod[1] and Williams, Ashleah[4]
[1]University of Exeter, [2]University of California, Davis, [3]University of Plymouth, [4]University of Queensland

Abstract: Wildfire frequency and magnitude have increased globally due to changes in land use, warming climate, and increased droughts. Subsequent rainfall washes ash into aquatic systems, exposing organisms to elevated pH, metal, and polycyclic aromatic hydrocarbon (PAH) levels – leading to mass mortality events. Past research found environmentally-relevant ash exposure caused up to 33% mortality in Chinook salmon (Oncorhynchus tshawytscha) despite individual components being magnitudes below reported LC50 values. Ongoing gill transcriptomics revealed what we suspected: ash-exposed salmon differentially expressed genes associated with base secretion (e.g. VHA-b, CFTR, CA) and metal ion removal (e.g. metalloproteinase, CNNM). While it is not surprising that a complex ash mixture is more toxic than individual components, we sought to dissociate the effects of alkaline water pH and contaminants (metals & PAHs) in two fish studies in the United Kingdom and Australia. Ash exposure (combined high pH & contaminant) caused significant mortality in rainbow trout (Oncorhynchus mykiss) alevin and silver perch (Bidyanus bidyanus); yet in all cases mortality was roughly halved when exposed to either high pH alone or the metals/PAH contaminants at control water pH. Surviving trout alevins exhibited no signs of delayed mortality, but did experience delayed somatic development. Trout alevin heart rate analysis is ongoing and will test whether ash exposure led to metabolic suppression. Altogether, our findings demonstrate the synergistic effects of alkaline water pH and contaminants – and that the removal of one of these components could be a viable method for preventing mass mortality events associated with wildfire runoff critical for conservation globally.

CHANGE II Investigating the interrelated responses of climate, contaminants, & biodiversity loss in fishes

Characteristics of drought in Amazonian-river systems: relevance to fish communities.

Johannsson, Ora[1] (*johannss@zoology.ubc.ca*) Oral Presentation
Co-Authors: Nascimento, Thiago[2]; Agasild, Helen[3]; Zingel, Priit[3]; Duarte, Rafael[2]; De Boeck, Gudrun[4]; Crémazy, Anne[5]; Morris, Carolyn[1]; Wood, Chris[1] and Val, Adalberto[6]
[1] The University of British Columbia; [2] Brazilian National Institute for Research of the Amazon; [3] Estonian University of Life Sciences, [4]University of Antwerp, [5]Institut National de la Recherche Scientifique, [6]Instituto Nacional de Pesquisas da Amazônia

Abstract: Drought and flood events are now more common in the Amazonian hydrological systems. These systems not only include the rivers but connecting channels and lakes, and the flooded forests. The more recent droughts of 2023 and 2024 are the worst on record. We investigated the conditions in two ecosystems at the nadir of the 2023 drought and interpret their effects on fish community 'health'. We sampled in the Anavilhanas Archipelago, a protected area in the Rio Negro (black water), and in lake Janauacá and the main river of the Rio Solimões system (white waters).
Water quality (pH, oxygen, temperature, conductivity, total suspended solids, dissolved organic carbon (DOC)) and zooplankton from near-surface waters were examined as well

as the microbial foodweb from integrated whole-water column samples.We saw marked decreases in fish habitats compared with past observations at both sites. Lakes were nearly gone in both systems.The water levels were shallow and in the Rio Negro suspended sediments were present even in near surface samples. Cladoceran abundance was negatively correlated with the quantity of larger suspended sediments. The chemical quality of the DOC was depressed and declined further in shallower areas. In the Rio Negro, very high temperature peaks were observed in the afternoon, while in the Rio Solimões bacteria overwhelmed the microbial foodweb. Potential and observed effects of these changes on the fish community together with literature information on the effects of extreme drought on fish community and composition will be discussed. (INCT ADAPTA III – CNPq, NSERC).

CHANGE II Investigating the interrelated responses of climate, contaminants, & biodiversity loss in fishes

Climate vs. Weather: Short-Term Environmental Change Leads to Altered Phenotypes Across Generations

Martinez Bautista, Gil (*gil.martinezbautista@unt.edu*) Oral Presentation
Co-Authors: Burggren, Warren
University of North Texas

Abstract: Many studies across fish biology focus on future effects of global climate change. However, the interpretation of 'climate change' has different implications for different ecosystems. Freshwater ecosystems are subject to environmental changes that can vary significantly over short periods of time in response to extreme weather events. Here we show examples of how short-term exposure to changes in temperature and dissolved oxygen modify fish morphological and physiological responses to these stressors, how they shape developmental trajectories and how they influence phenotypes in subsequent generations. We show how juvenile, male, and female Siamese fighting fish (Betta splendens) and blue gourami (Trichopodus trichopterus) adjust their ventilatory rates as a function of acute changes in temperature and progressive hypoxia that might mimic weather events. Also, we describe how changes in temperature and hypoxia modulate growth rate, metabolic rate, and cardiac function in Zebrafish (Danio rerio) during early development. Also affected are the dynamics of inheritance of hypoxia-tolerant phenotypes across clutches within a single generation or across multiple generations after short-term parental exposure to hypoxia. We show that different phenotypes emerge depending on the type of stressor, the dose of a given stressor, and the time of exposure. These differences occur within experimental groups, across development, and between sexes in adult fish. Additionally, some of these modifications last across generations. Collectively, our data suggest that conditions mimicking short-term weather-driven environmental change have profound implications for fish performance, from rapid physiological adjustments to long-term modifications that may affect species- and populational level survival.

CHANGE II Investigating the interrelated responses of climate, contaminants, & biodiversity loss in fishes

Individual and cumulative effects of nylon microplastic fibres and warming temperature on threespine stickleback (Gasterosteus aculeatus) behavior

Hajji, Angelina Lyla (*angelina.hajji@ucalgary.ca*) Oral Presentation
Co-Authors: Lucas, Kelsey
University of Calgary

Abstract: In Canada, climate is warming at twice the global rate; as the Canadian Pacific absorbs the heat, marine temperature regimes are starkly impacted and consequently so are fishes. Concurrently, plastics, crucially nylon used in fishing industries, are contaminating marine waters at unprecedented levels, with detrimental effects on fishes. Despite populations being exposed to multiple stressors simultaneously, cumulative effects remain poorly understood. Here we investigated the cumulative effects of warming and nylon microplastic fibres on the feeding behavior of threespine stickleback (Gasterosteus aculeatus) by exposing fish to conditions of 15°C and 20°C and nylon concentrations of 0, 1, 10, and 100 mg/g (mg nylon/g food) for 4 weeks. Feeding rates responded complexly to multiple stressors, demonstrating the highest rates under warming conditions while increasing concentrations of plastic reduced feeding rates. In response to microfibre ingestion, we observed “coughing” behaviors, and a unique reselection tendency of food items selected by conspecifics. Given these results, we anticipate greater anxiety, impeded conspecific cueing, reductions in foraging efficiencies with increasing plastic concentration. Cumulatively, these stressors will yield greater energetic trade-offs and decreased accuracy in food selection with stark implications for ecosystem dynamics.

CHANGE II Investigating the interrelated responses of climate, contaminants, & biodiversity loss in fishes

Caught in a thermal loop: Unraveling the molecular, biochemical, and whole-animal responses to diel thermal variability in zebrafish (Danio rerio)

Clow, Tanner (*tclow@uwaterloo.ca*) Oral Presentation
Co-Authors: Craig, Paul
University of Waterloo

Abstract: Aquatic organisms live in complex, dynamic environments, where a myriad of abiotic factors, most notably temperature and dissolved oxygen, undergo daily fluctuations. Due to their inherent relationship, these abiotic factors are thought to share common signalling pathways (cross-talk) and protective mechanisms (cross-tolerance). However, despite temperature being one of the most influential abiotic factors, much of the current knowledge of its effects on fish stems from static exposures. Therefore, in this study, we took a holistic approach and examined the effects of thermal variability across multiple levels of biological organization, ranging from molecular to whole-animal. We hypothesized that diel temperature fluctuations would alter whole-animal thermal and hypoxia tolerance through various underlying cellular mechanisms involved in cross-talk

and cross-tolerance. To address this hypothesis, we acclimated adult zebrafish to either control conditions (27ºC) or a thermal flux (23-33ºC) for approximately two weeks. Following the acclimation, we quantified whole-animal thermal (critical thermal maximum) and hypoxia (time to loss of equilibrium) tolerance, along with several heat-shock response genes, hypoxia response genes, microRNAs (miRNAs), heat-shock proteins, and metabolic enzymes in the brain and liver tissue. We demonstrated that when acclimated to diel temperatures, zebrafish are better equipped to withstand acute thermal and hypoxia challenges. Accompanying this change in whole-animal tolerance, we observed differential expression of miRNAs, mRNAs, proteins, and enzyme activity between static and fluctuating conditions. Taken together, these findings strengthen our collective understanding of the physiological consequences associated with thermal variability.

CHANGE II Investigating the interrelated responses of climate, contaminants, & biodiversity loss in fishes

Stress in the waters: Physiological and morphological responses to environmental stressors in imperilled fishes from Southwestern Ontario

Dodsworth, Emily[1] (*e2dodswo@uwaterloo.ca*) Oral Presentation
Co-Authors: Craig, Paul[1] and Firth, Britney[2]
[1]University of Waterloo, [2]University of Alberta

Abstract: In Southwestern Ontario, anthropogenic activity, such as agriculture and urbanization, heavily influence aquatic ecosystems and the local fish populations which inhabit them. These aquatic habitats can be disturbed by surface runoff resulting in an influx of various contaminants, nutrients, and sediment loadings that leach into water bodies altering their water chemistry, temperature, and natural processes. As ectotherms, fish experience body temperatures that closely track ambient conditions. Consequently, changes in the environment directly influence physiological processes, meaning environmental stressors can profoundly affect metabolism, thermal tolerance, behaviour, and ultimately fitness. In our study, we investigated how different environmental stressors effect the physiology and morphology of three At-Risk fish species: The blackstripe topminnow (Fundulus notatus), the eastern sand darter (Ammocrypta pellucida), and the redside dace (Clinostomus elongatus). This was accomplished by sampling from both urban and rural populations which experienced different temperature profiles, sediment loadings, and contaminant concentrations. At each sampling site, respirometry and critical thermal maximum (CTmax) tests were conducted to determine aerobic scope and thermal tolerance, respectively, which was then compared between populations. For F. notatus, laboratory tests were also performed, focusing on chronic turbidity exposure and its impact on gill morphology and behaviour. Results demonstrated that thermal tolerance increased with acclimation temperature, while aerobic scope remained relatively consistent among populations of the same species, despite differences in local thermal regimes and contaminant exposure. Collectively, these findings address how multiple environmental stressors shape physiological performance, informing conservation strategies and

predictions about the long-term persistence of imperilled species under changing environmental conditions.

CHANGE II Investigating the interrelated responses of climate, contaminants, & biodiversity loss in fishes

Decadal trends in metal contamination and fish community responses in an industrialized river system (Nete river basin, Flanders, Belgium)

Balthazar, Iris[1] (*iris.balthazar@uantwerp.be*) Poster Presentation
Co-Authors: Teunen, Lies[2] and Blust, Ronny[1]
[1]University of Antwerp; [2] INBO, Research Institute for Nature and Forest, Antwerp

Abstract: European riverine ecosystems face mounting pressures from anthropogenically induced habitat degradation and water quality deterioration, with fish communities serving as sensitive indicators of ecological health. Industrial and agricultural activities along rivers can significantly alter water chemistry, particularly through metal contamination, with cascading effects on fish populations and biodiversity. This study investigates the long-term (2013–2023) dynamics of metal concentrations, water quality, and fish community structure in the Nete river basin, a river system in Flanders, Belgium, characterized by intensive industrial activity.

Leveraging an extensive monitoring network established by the Flanders Environment Agency, we analyzed temporal trends in metal concentrations and water quality parameters, alongside fish stock assessments performed by the Research Institute for Nature and Forest, using the Ecological Quality Ratio (EQR) derived from the Flemish Index of Biotic Integrity (IBI). The IBI, aligned with the European Water Framework Directive, provides a robust framework for evaluating riverine ecological status based on fish community data collected via standardized electrofishing surveys in bream and barbel zones.

Our analysis looks into correlations between metal contamination, water quality, and fish community metrics, taking into account shifts corresponding to changes in local industrial activity. These findings underscore the interconnected impacts of contaminant exposure and habitat quality on fish biodiversity, offering critical insights for managing industrialized river systems in the face of climate change and ongoing anthropogenic pressures.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

Gary Anderson, Ian Bouyoucos and Alyssa Weinrauch

Abstract: Agnathans (i.e., hagfish and lampreys) and chondrichthyans (i.e., sharks and rays) are some of the primitive fishes that are useful for investigations of the evolution of physiological systems. Agnathans diverged from the vertebrate lineage after the first whole genome duplication, while chondrichthyans diverged after the second, yet prior to the within lineage duplications occurring in the more derived actinopterygians. Thus, these enigmatic groups of fishes are often described as 'living fossils' and have been the subject of studies of comparative physiology for decades, both from a fundamental evolutionary perspective and in the context of environmental change. This symposium examines advances in the field in both contexts from across an international stage.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

Agnathans and Chondrichthyans as models for digestive physiology

Weinrauch, Alyssa (*alyssa.weinrauch@ubc.ca*) Oral Presentation
The University of British Columbia

Abstract: Agnathans and Chondrichthyans represent the earliest diverging extant vertebrate groups making them useful for the study of functional evolution of many physiological systems. This is particularly true for studies of the gastroenteropancreatic system as there is a prominent morphological transition from a multifunctional tube that performs each of these roles in agnathans, to discrete and compartmentalized organs with specialized roles in chondrichthyans. Further, this organ system is crucial to regulate metabolic homeostasis in the face of changing energetic needs across postprandial stages. Indeed, the emergence of a discrete pancreas represents a significant evolutionary advancement that is hypothesized to permit evolution of more complex gut-organ axes, perhaps through the heightened control of plasma metabolites. Aspects of exocrine and endocrine pancreatic function have been investigated in the hagfishes and elasmobranchs, yet much remains to be determined regarding stimuli, timing, magnitude of response, and mechanisms controlling secretion. In addition, aspects of environment and life history must be considered given the unique metabolism and feeding habits, including spatially and temporally separated meals, of these taxa. Ultimately, interpretation of physiological data are complicated by whether these traits are derived or primitive in these charismatic fishes.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

The latest research on spiny dogfish (Squalus acanthias) aquaporin gene/protein expression…

Cutler, Christopher (*ccutler@georgiasouthern.edu*) Oral Presentation
Georgia Southern University

Abstract: In previous ICBF presentations, details of AQP10 paralogs were presented. Further research has been ongoing concerning these genes/proteins. RT-PCR expression of the AQP10C1 full length coding region showed that full length transcripts are only found in the gill. Previously, AQP10C2 expression was shown to be localized to various cells in the intestinal tract which were identified as likely to be neuroendocrine cells. Quantitative PCR experiments were conducted which showed the expression of AQP10C2 was very low and yet the antibody staining was strong. So, a second AQP10C2 antibody was made to attempt to confirm the results. The second antibody colocalized in the same cells as the first AQP10C2 antibody, but the subcellular distribution of staining was different, suggesting post-translational modification of the AQP10C2 protein resulting in blockage of antibody binding sites. Further work on Aquaporin gene/mRNA splice variant forms may also be presented time allowing.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

Dietary state influences olfactory and cardiovascular responses to odorants in North Pacific Spiny Dogfish (Squalus suckleyi)

Edwards, Tyler[1] (*edward32@myumanitoba.ca*) Oral Presentation
Co-Authors: Fry, Mark[1]; Anderson, W. Gary[1]
[1]University of Manitoba

Abstract: Olfaction is a key sensory system in animals and plays critical roles in feeding, predator avoidance and conspecific communication. In fishes, odours are detected by receptors of olfactory sensory neurons (OSNs) located in the olfactory epithelium, initiating integrated physiological responses that vary with odorant and dietary state. OSNs are bipolar neurons whose apical dendrites extend into the external environment, while their axons project to the olfactory bulb. From the olfactory bulb, signals are relayed via synapse to higher neural centers involved in feeding regulation, including the telencephalon and hypothalamus. Hypothalamic nuclei further project to brainstem regions such as the medulla oblongata which regulates autonomic functions including heart rate. The goal of this study was to investigate the physiological changes in odorant detection and the integration of these signals by the cardiovascular system in North Pacific Spiny Dogfish (Squalus suckleyi) under different dietary states. Olfactory responses were assessed using electro-olfactogram (EOG) technique in fed and fasted (+7-day) individuals by measuring responses to varying concentrations of L-alanine (n=6/treatment). Cardiovascular

responses to dietary (squid, L-alanine), environmental (bile) odorants and a control (sea water) were recorded in fed (n=5-6) and fasted (n=8) free-swimming individuals. Fasted individuals exhibited significantly increased EOG responses to L-alanine and a pronounced cardiovascular response to squid. These findings provide new insight into olfactory–autonomic integration in elasmobranchs and highlight the influence of dietary state on sensory and physiological processing.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

Impacts of warming on cardiac function in the temperate Pacific spiny dogfish (Squalus suckleyi).

Folkerts, Erik[1] (*Erik.Folkerts@umanitoba.ca*) Oral Presentation
Co-Authors: Anderson, W. Gary[2]
[1]University of British Columbia; [2]University of Manitoba

Abstract: The Pacific Northwest is a region experiencing robust changes in many coastal and non-coastal bodies of water. Although buffered by a larger volume, more frequent and severe changes in coastal water temperature are being observed, with devastating impacts on marine wildlife. Although some investigation on thermal performance has been previously made, many unknowns and specifics of cardiac thermal preference in the Pacific spiny dogfish (Squalus suckleyi) remain under-investigated. Cardiac thermal performance was evaluated in this species using both in vitro and in vivo techniques. Utilizing an organ bath apparatus, cardiac contractile properties were assayed under varying temperature regimens (static versus thermal ramp profiles) to gauge tissue-level thermal limits of contraction frequency, speed, magnitude, and sensitivity. Cardiac Ca2+ cycling under such temperature profiles was also examined by application of specific Ca2+ channel pharmacological blockers and manipulated bath water chemistries. At the level of the whole organism, fish were subjected to acute increases in temperature and monitored for changes in cardiac output (via surgically implemented coronary flow probes). Dorsal aorta cannulations also allowed simultaneous recordings of blood pressure during acute warming stresses. Collectively, our combined in vitro and in vivo analyses allowed us to refine Pacific spiny dogfish thermal cardiac limits, as well as make novel discoveries concerning physiological responses to temperature stress. Considering its economic importance and trophic positioning in many food webs in the Pacific Northwest region, our results contribute directly to a better understanding of how this vital chondrichthyan may cope under changing future conditions.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

Molecular characterization of potassium channels and their osmoregulatory role in the gills of sea lamprey

Cannizzaro, Ava (*ava.cannizzaro@okstate.edu*) Oral Presentation
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Abstract: Vertebrate K^+ channels belong to a large evolutionary conserved yet structurally diverse gene family that regulates fundamental cellular processes, including an essential role in maintaining membrane potential, which is particularly critical in excitable and ionoregulatory cell types. Sea lamprey are the most basal vertebrate capable of osmoregulation and undergo a larvae-to-juvenile metamorphosis during which they develop the ability to survive the transition from freshwater (FW) to seawater (SW). Thus, sea lamprey serve as a good model and useful evolutionary comparison in investigations into the role of K^+ channels in osmoregulation in fishes. This study aimed to identify and characterize the role of K^+ channels in the gills of sea lamprey that support osmoregulation in FW and SW. We first identified all K+ channel genes present in sea lamprey by analyzing the recently released assembly of the sea lamprey genome. We used molecular evolutionary analyses to determine the putative identity of these genes in relation to the broader K+ channel gene family that emerged during the radiation of vertebrates.We compared gene expression of K+ channels between FW larvae, FW juveniles, and SW juveniles. We further analyzed the entire branchial transcriptomes of FW larvae and FW juveniles. Findings from these analyses revealed significant upregulation of two members of the inwardly rectifying K+ channel (Kir) family during metamorphosis and SW acclimation, with strong evidence supporting Kir4 (kcnj10/15-like) as the primary candidate gene for supporting branchial ionoregulation in SW. Thus, the involvement of Kir appears to be a deeply conserved mechanism among salt secreting ionoctyes.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

Anoctamin1/TMEM16A expression in the spiny dogfish (Squalus acanthias): Is one of its roles likely to be osmoregulatory?

Cutler, Christopher (*ccutler@georgiasouthern.edu*) Oral Presentation
Georgia Southern University

Abstract: Anoctamin1/TMEM16A (ANO1) is known to be a calcium-activated chloride channel (CACC) that is fairly ubiquitously expressed in mammalian tissues where the gene possesses three splice variant versions. Recent research on teleost fish ANO1 suggests that as well as a role in tissues such as muscle, it may have an osmoregulatory role in chloride secretion in the gill. Hence ANO1 had the potential to play an osmoregulatory role in elasmobranchs. The spiny dogfish ANO1 cDNA/mRNA coding region was amplified by nucleotide degenerate RT-PCR and as well as the main coding sequence, four splice variant versions were identified. Amplification of the coding region was seen in 7 out of 10

tissues including rectal gland, kidney and intestine but not the gill. A custom antibody was made against a peptide from the C-terminal region of the derived ANO1 protein sequence. In the intestine, ANO1 cell localizations showed some similarity to those concerning Aquaporin 10C2. Localization of the ANO1 protein to date though suggests the lack of an osmoregulatory role in sharks. Further ongoing intestinal colocalization results will be presented if successful.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

Effects of salinity transfer on cardiac function in the partially euryhaline North Pacific spiny dogfish (Squalus suckleyi)

Hutton, Melody (*melody.hutton@gmail.com*) Oral Presentation
Co-Authors: Anderson, W. Gary
University of Manitoba

Abstract: Nitrogen is an essential element for all eukaryotic organisms, contributing to amino acids, DNA, and proteins. Elasmobranchs (sharks, rays) are among the most ancient vertebrates and uniquely retain high concentrations of the nitrogenous osmolyte urea to maintain iso- or hyperosmotic balance in marine habitats. Fully euryhaline, and to a lesser extent partially euryhaline, species adjust plasma sodium, chloride, trimethylamine oxide (TMAO) and urea concentrations to regulate intra- and extracellular osmolality following changes in environmental salinity. In addition to osmoregulation, elasmobranch cellular metabolism relies on nitrogen as amino acids and ketones serve as primary metabolic fuel, with glucose playing a more variable, tissue-specific role. Therefore, osmoregulatory and metabolic processes may be linked as nitrogen is required for both urea synthesis and metabolic fuel use. Salinity-driven responses in the gills, intestine, liver, and kidneys are well documented; however, effects on cardiac tissue remain largely unexplored. Given that salinity may alter blood volume and osmolyte concentrations, I hypothesize that environmental salinity changes impact cardiac performance and metabolism in spiny dogfish. Specifically, this study assesses shifts in cardiac metabolic fuel use (amino acids, glucose, ketones) and determines how salinity acclimation affects heart morphology and contractility. Results indicate a metabolic shift toward greater glucose reliance in high-salinity-acclimated dogfish, along with tissue-level changes in cardiac contractility in both low- and high-salinity treatments. Together, these findings clarify how the heart responds to salinity acclimation in partially euryhaline sharks and whether shared nitrogen demands integrate metabolic and osmoregulatory processes to maintain cardiac function in an ancient vertebrate lineage.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

Blueprints for survival: Genetic mechanisms underpinning physiological performance in the epaulette shark Hemiscyllium ocellatum

Hasenei, Aaron[1] (*aaronhasenei@gmail.com*) Oral Presentation
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Abstract: As an early-diverging vertebrate lineage, Chondrichthyan fishes offer valuable insight into the evolutionary foundations of physiological regulation. This study uses the epaulette shark (Hemiscylliumocellatum) as a model system to examine how geographic variation and thermal history shape intraspecific limits of performance under ocean warming scenarios. Using acute and chronic warming experiments across three genetically distinct populations spanning nearly 7° of latitude, we found that the low- and mid-latitude populations maintained aerobic scope, oxygen transport efficiency, and enzyme activity up to 32°C. In contrast, the high latitude population evinced a 32% decrease in aerobic scope, driven by a drastically elevated standard metabolic rate (Q10 = 4.6), and a reliance on anaerobic pathways. Complementing these results, differential gene expression profiles provided evidence of local thermal specialisation; however, the capacity for phenotypic plasticity was not universal, with clear implications for survivability. These findings show co-gradient responses, where low latitude populations exhibit greater thermal resilience through their higher capacity for genetic and physiological phenotypic plasticity in contrast to their high latitude conspecifics that exhibit heightened thermal vulnerability. Key genetic drivers identified within the differential gene expression analysis were mechanistically linked to the physiological performance phenotypes using weighted gene co-expression network analysis to ascertain integral molecular pathways that contribute to survival or vulnerability to heat stress. By linking population-level genetic variation with organismal performance, this work provides new insight into how chondrichthyan fishes may respond to ongoing ocean warming and highlights the value of ancient vertebrate lineages for understanding the evolution of physiological resilience.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

An integrative approach toward quantifying physiological responses to stressors in spiny dogfish

Debaere, Shamil[1] (*shamil.debaere@uantwerpen.be*) Oral Presentation
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Abstract: The physiological correlates of prolonged, repeated stressor exposure in elasmobranch fishes remain poorly explored. Using an integrative approach, by linking blood chemistry, gene expression, heart rate, and aerobic performance, weset out to quantify responses to acute and prolonged stressors in a representative elasmobranch species, the spiny dogfish (Squalus suckleyi). We surgically implanted heart rate loggers in

the abdominal cavity of the dogfish, and following a 5-day recovery period, we applied a daily stress paradigm to experimentally simulate exposure to repeated stressors over a three-week experiment. Once every week, the dogfish were sampled by caudal venipuncture. One of two treatments was subsequently confined in individual 50L blacked-out isolation boxes for 48h (during which oxygen consumption rates were monitored); the other treatment was returned to its original 150.000L holding tank. Basal plasma glucose and β-hydroxybutyrate (β-HB) concentrations of a third control group (that did not undergo surgery) remained stable and around baseline values from the wild, whereas plasma glucose and β-HB increased significantly in both treatments that underwent surgery. These changes in plasma glucose and β-HB coincided with similar changes in transcript abundances of glucose and monocarboxylate transporters in liver and white muscle tissues. Additionally, heart rates peaked on sampling days (following an acute stressor) and remained consistently elevated by the end of the experiment (following prolonged stressor exposure). Oxygen consumption rates at rest and after exhaustive exercise, however, did not change throughout the experiment. These data provide a new, broad look at the suite of physiological responses to stressors in this representative elasmobranch species.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

How Pacific Hagfish (Eptatretus stoutii) Survive Reoxygenation Following Extended Anoxia

Yutsyschyna, Maria[1] (*myusishe@uoguelph.ca*) Oral Presentation
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Abstract: Pacific hagfish (Eptatretus stoutii) are an agnathan fish with the ability to tolerate 36 hours of anoxia exposure. Previous studies indicate that this species meets the challenge of energy limitation during sustained anoxia with a moderate reduction in their already low metabolic rate allowing their energy requirements to be met via glycolysis alone. However, other challenges of anoxia manifest upon reoxygenation. Mitochondrial dysfunction can produce a surge of reactive oxygen species (ROS) causing widespread oxidative damage. We investigated how Pacific hagfish survive reoxygenation stress by examining the accumulation of succinate during anoxia which can drive mitochondrial dysfunction, the upregulation of antioxidant enzymes, and the appearance of oxidative damage biomarkers. Succinate accumulated 9-fold in the heart and 3.5-fold in the muscle following 24 hours of anoxia. Less than the accumulation in anoxia intolerant animals. The activity of the antioxidant enzyme catalase in the liver was unchanged following 30 hours of anoxia but was upregulated 4- and 5-fold after 1 and 3 hours of reoxygenation. Catalase was increased 3-fold in the muscle after 3 hours. Catalase activity had returned to baseline in both tissues after 6 hours. Protein carbonyls, a biomarker of oxidative damage, were elevated in the liver following 3 hours of reoxygenation and had recovered after 6 hours. These results indicate that Pacific hagfish can reduce the driving force of mitochondrial dysfunction and upregulate antioxidant activity to respond to the increased oxidative stress

of reoxygenation. What oxidative damage that occurs is minor and fully recovered within 6 hours of reoxygenation.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

Physiological Performance and Stress Resilience in Larval Sea Lamprey.

Wilkie, Michael[1] (*mwilkie@wlu.ca*) Oral Presentation
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Abstract: The sea lamprey life cycle includes a prolonged (3–7 year) burrow-dwelling larval stage, followed by a profound metamorphosis characterized by formation of an oral disc and dagger-like tongue which are used to ingest the blood of teleost and chondrichthyan fishes. There remain gaps, however, in our understanding of how lampreys respond to environmental stressors. We examined the burrowing performance of larval sea lamprey, as well as their ability to recover from an acute stressor. Using a custom "burrowing performance chamber", we assessed the metabolic costs of burrowing. Larval lamprey became completely burrowed within 11 seconds, but the metabolic costs were only moderate and short-lived, as indicated by a relatively small increase in muscle lactate. Muscle ATP was sustained by phosphocreatine, which decreased by 50% but was rapidly restored. Exposure to a standard stressor (chasing for 5min plus 1min netting) led to a rapid, but relatively modest 3-fold elevation of the corticosteroid, 11-deoxycortisol, and 3–4 fold increases in plasma and liver glucose, with no change in liver, brain, or muscle glycogen. Moreover, patterns of liver mRNA expression for key stress markers (e.g., corticosteroid receptor, gluconeogenic enzymes), measured with a novel lamprey high-throughput qPCR stress chip, were similar in stressed lamprey and unstressed control lamprey. We conclude that larval sea lamprey possess metabolic adaptations that facilitate rapid energy mobilization for critical behaviours such as burrowing or in response to acute stressors. Recovery from burrowing or acute stressors is also rapid, likely reflecting a high degree of physiological resilience during this life stage of the sea lamprey life cycle.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

Regulatory mechanisms of selective follicle development, maturation and ovulation leading to an ovarian hierarchy in cloudy catshark

Inoue, Ryotaro[1] (*inoue@aori.u-tokyo.ac.jp*) Oral Presentation
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Abstract: In contrast to teleosts, chondrichthyans (sharks, rays, and chimaeras) exhibit unique reproductive strategies. Over half of all species are viviparous, and all copulate. Females often store sperm and reproduce multiple times without mating. We recently reported that follicles of different developmental stages are present in the ovary of oviparous cloudy catshark, Scyliorhinus torazame, comprising a consistent hierarchy of two vitellogenic follicles at each stage (e.g. from the largest, two F1 follicles, two F2 follicles, and so on). Following the plasma luteinizing hormone (LH) surge, F1 follicle pairs produce progesterone (P4) through upregulation of steroidogenic acute regulatory protein 2 (star2) and downregulation of 17α-hydroxylase/17,20 lyase (cyp17α1), which drives the plasma P4 surge preceding to ovulation. On the other hand, regardless of LH receptor (lhr) expression, the LH surge did not induce star2 expression in F2 and smaller follicles, and failed to stimulate P4 production in these follicles, despite a concurrent testosterone reduction which may be the additional function of LH. We further established in vitro follicle culture system and found that pituitary ventral lobe extract induced star2 expression exclusively in F1 follicle and upregulated the expression of multiple genes associated with ovulation. Although both F1 and F2 express lhr, the mechanism underlying their different responses to the LH surge remains unclear; nevertheless, we propose the cloudy catshark as a suitable model for characterizing chondrichthyan reproductive physiology and studying the regulatory and evolutionary aspects of hierarchical selection of ovarian follicles in vertebrates.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

Sharks, steroids, and symbiosis? An investigation into the biosynthetic pathway of 1-alpha-hydroxycorticosterone

Cederberg, Elliot[1] (*cederbee@myumanitoba.ca*) Poster Presentation
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Abstract: Corticosteroids are produced in the adrenal/interrenal gland through an enzyme-regulated pathway, and display either glucocorticoid (energy metabolism) or mineralocorticoid (electrolyte balance) roles. While the process of corticosteroid synthesis has been well described in tetrapods and teleosts, it remains largely undescribed for the primary elasmobranch (sharks/rays) corticosteroid; 1-alpha-hydroxycorticosterone (1a-OHB). Interestingly, in the original identification of 1a-OHB, it was proposed that the unique configuration of this steroid hormone could be due to microbial transformations, which aligns with our findings of the presence and identification of bacteria in healthy elasmobranch interrenal tissues. My current research focuses on decoding the biosynthetic pathway of 1a-OHB in the Pacific spiny dogfish, Squalus suckleyi, through the identification of steroid intermediates, enzymatic activity, and possible microbial interactions within this process. Identification of steroid intermediates will be revealed using analytical chemistry techniques (HPLC, NMR). Identification of intermediates will be coupled with measuring the transcript abundance of steroidogenic enzymes involved in

this pathway, with the help of a developed transcriptome of the interrenal gland of S. suckleyi. Lastly, the potential role of bacteria in mediating steroid transformations will be examined using established incubation protocols with recently isolated bacterial strains from the interrenal gland of S. suckleyi. This will be done in combination with sequencing and fluorescent in situ hybridization (F.I.S.H), allowing the localization of bacteria within the interrenal gland. Understanding the complete biosynthetic pathway of 1a-OHB will address the role/importance of this unusual steroid and may also reveal a novel host-microbiome relationship in vertebrates, specifically in the context of corticosteroidogenesis.

ANCIENT Ancient fishes in comparative physiology: Agnathan and chondrichthyan fishes

Relaxed purifying selection in stress axis genes of ancient fishes relative to teleosts

Lazaro-Côté, Analisa[1] *a.lazarocote@gmail.com* Poster Presentation
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Abstract: The neuroendocrine stress response is well-conserved across vertebrates, playing an important role in maintaining homeostasis upon stressor perception. The hypothalamic-pituitary-interrenal (HPI) axis is particularly well-studied in teleosts, but less in known in Acipenseridae (sturgeons and paddlefish) and Elasmobranchii (sharks and rays). Functional divergence among these groups may drive selection pressures on protein-coding genes involved in the neuroendocrine stress response. Proper functioning of the HPI axis is critical in teleosts, therefore we hypothesized that extensive purifying selection maintains the function of HPI-related genes in teleosts. By contrast, Acipenseridae and Elasmobranchii may rely on compensatory or alternative pathways for stress responses, which could result in relaxed purifying selection on the HPI axis relative to teleosts. Therefore, we analyzed whole genomes of Acipenseridae (n = 5), Elasmobranchii (n = 7), Teleostei (n = 8), and outgroups (n = 9), for ortholog identification, multiple sequence alignment, and HyPhy RELAX to test for selection intensity. We curated a set of genes involved in HPI axis function in teleosts and included 102 randomly selected genes to infer genome-wide patterns of selection. We found evidence for relaxed purifying selection in genes of the HPI axis in Acipenseridae and Elasmobranchii, in addition to genome-wide relaxed selection. Our results suggest that although this relaxed purifying selection largely reflects genome-wide evolutionary forces, functional divergence between ancient fishes and teleosts may drive relaxed selection in specific elements of the neuroendocrine stress response.

MOTION I Metabolism in motion: Mechanics, physiology, and ecology of fish movement

Metabolism in motion: Mechanics, physiology, and ecology of fish movement

Yangfan Zhang, Katja Anttila and George Lauder

Abstract: Understanding fish movement is a central challenge in biology that integrates decades of fundamental research in physiology, biomechanics, energetics, and hydrodynamics. We are now at the edge of a profound transformation. Emerging and rapidly maturing tools, such as AI-enhanced kinematic tracking, minimally invasive biologgers on free-moving animals, robotic surrogates, and ocean-scale tracking, are shifting the paradigm: from individual to collective behaviour, from the energetics of fine-scale kinematics to activities of large-scale movement, and from controlled experiments to real-world complexity. The relevant topics in this symposium include, but are not limited to, kinematic-metabolic coupling, collective movement, climate driven range shifts, as well as physiological limits of fish locomotion, and their implications for conservation, aquaculture, and welfare, by asking how fish generate, regulate, and optimize movement and/or energetics in dynamic environments.

MOTION I Metabolism in motion: Mechanics, physiology, and ecology of fish movement

Linking energetics, acoustic telemetry, hydrodynamics, and neurophysiology to understand wild fish behavior

Liao, James (*jliao@ufl.edu*) Oral Presentation
University of Florida

Abstract: Understanding the mechanisms underlying fish locomotion requires studying behavior in ecologically relevant contexts. Although controlled experiments provide high-resolution biomechanical and physiological measurements, it remains unclear how well these findings generalize to natural environments. Here we developed a multi-scale framework that integrates field telemetry, mesocosm-based behavioral quantification, laboratory respirometry, and emerging in vivo neurophysiology to evaluate the drivers of movement in the wild.

At the landscape scale, acoustic telemetry of 43 red drum (Sciaenops ocellatus) across 5,610 km^2 of coastal Florida revealed marked individual variation in seasonal migration and tidal coupling, highlighting context-dependent behavioral strategies. At intermediate scales, dual tri-axial accelerometers affixed to the tail and operculum of red drum in a large outdoor mesocosm resolved swimming, resting, and maneuvering behaviors that are often indistinguishable using traditional dynamic body acceleration metrics. Parallel laboratory respirometry experiments provide energetic baselines that enable comparison between laboratory swimming performance and free-ranging movement patterns.

To probe neural mechanisms, we have developed a waterproof head-stage amplifier for real-time electrophysiological recordings in freely swimming fishes, laying the groundwork

for linking brain activity to natural locomotor behavior. Together, these approaches begin to quantify the relative contributions of energetics, sensory inputs, and environmental variables in shaping movement decisions.
By connecting environmental context, physiology, and neural state, this framework provides both a critical test of whether laboratory-derived results accurately reflect ecological reality and advances new mechanistic understanding of behaviors in nature.

MOTION I Metabolism in motion: Mechanics, physiology, and ecology of fish movement

An optimised workflow for automated multiple fish tracking in challenging experimental settings

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Automated animal tracking has greatly expanded opportunities to study behaviour through high-resolution, quantitative analysis of movement in space and time. Idtracker.ai (previously idTracker), developed by G. Polavieja and collaborators, represented a major advance in this field by enabling the non-invasive tracking of unmarked individuals based on their unique visual features. Beyond reliable identity assignment, the software offers tools for trajectory validation and correction and can support the matching of individuals across tracking sessions, thereby increasing both analytical flexibility and data quality. Despite these advances, tools like idtracking.ai are often underused, particularly where recording conditions are suboptimal, where reflections, low contrast, and occlusions can compromise identity preservation and trajectory quality. At the same time, the analysis of the trajectories output is not often time-consuming.

Here, we present an optimised workflow for fish automated tracking that integrates idtracker.ai with a practical preprocessing and annotation pipeline designed for difficult experimental environments. The workflow combines Detectron2-based segmentation with LabelMe-assisted annotation and correction to improve animal detection in low-quality or complex footage. Through iterative training, targeted frame selection, and protocol-based refinement of annotations, the pipeline increases tracking robustness while retaining flexibility across setups, supporting the extraction of high-quality trajectories that can be used for downstream behavioural analyses at both the individual and group level. Beyond presenting a practical solution to a widespread technical bottleneck, this work aims to lower the entry barrier for advanced trajectory-based behavioural analysis in fish research. Ongoing development of a user-friendly downstream analysis tool will further extend this framework, with the goal of making sophisticated tracking and locomotion analytics more accessible, reproducible, and broadly adopted across the community.

MOTION I Metabolism in motion: Mechanics, physiology, and ecology of fish movement

The effect of salinity on the hypoxia tolerance, metabolism and swimming performance of a euryhaline teleost, white perch Morone americana

Nelson, Jay (*jnelson@towson.edu*) Oral Presentation
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Abstract: Hypoxia in estuaries like Chesapeake Bay has increased recently due to cultural eutrophication and climate warming. The ability to cope with hypoxic zones that shift through the water column may be critical for the Darwinian fitness of some estuarine fishes. White perch (Morone americana) occupy many salinity habitats and thus may be exposed to hypoxia at different salinities, yet whether hypoxia tolerance (HT) is salinity dependent is poorly known. We acclimated white perch to 0‰, 5‰, and 10‰ and subjected them to two hypoxia challenge tests (HCT) and two tests of swimming performance (Ucrit with metabolic rate and sprint) over an eight month period. Increased salinity negatively affected fish growth; fish acclimated to 10‰ had significantly slower growth than those in freshwater. Critical swimming (Ucrit) performance also decreased with increased salinity but not significantly so. There was no effect of salinity on sprinting ability. Salinity also did not affect HT, but its influence may have been obscured by a large time effect. A fish's second HCT was performed at various time intervals from their first; HT approximately doubled after an initial HCT if re-measured within 50 days, similar to congeneric striped bass, but then faded back towards original HT if measured after two months. Relative heart and gill size after 8 months was independent of salinity. Much of the individual variance in performance was due to "athletic" differences amongst these fish as HT was significantly correlated with both Ucrit and sprint swimming performances, i.e. better swimmers were more hypoxia tolerant.

MOTION I Metabolism in motion: Mechanics, physiology, and ecology of fish movement

Survival of the slowest? Using activity to predict climate change vulnerability of fishes

Dichiera, Angelina (*dichiera@vims.edu*) Oral Presentation
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Abstract: For many fishes, the combined stressors of warming and deoxygenation are intricately linked by the dynamic balance between oxygen supply and demand. Increases to the minimal energy requirements for survival and any constraints to the energy available for activity beyond the cost of living (e.g., growth, reproduction) will negatively impact a fish's ability to tolerate stressors. Because this supply and demand is inextricably linked to activity, diversity in lifestyle – whether fish is more sedentary, more active, or anywhere in between – may provide insight into which organisms are more vulnerable to co-occurring stressors. In other words, activity level may allow us to predict the "winners" and "losers" in future climate change scenarios. To determine if there is a relationship between activity

and environmental stress tolerance, eight local estuarine fish species were characterized for their vulnerability to temperature stress using critical thermal maxima (CTmax). Six of these species were additionally tested for CTmax in combination with decreased oxygen availability. Morphometric indices were used to predict swimming capability and tissue samples were taken to measure capacity for aerobic and anaerobic metabolism. Low-activity species are predicted to be more resilient than high-activity species and may have more energetic stores to supplement aerobic metabolism in oxygen-limited environments. Should there be no support for a predictive relationship between vulnerability and activity as hypothesized, we expect these data will still offer meaningful avenues for identifying other factors that shape which species will be climate change "winners" or "losers".

MOTION I Metabolism in motion: Mechanics, physiology, and ecology of fish movement

Exercise training in rainbow trout (Oncorhynchus mykiss): from cardiac action potential modulation to heart rate adjustments

Le, Thuy[1] (*thuy.le@utu.fi*) Oral Presentation
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Abstract: Exercise training is known to improve cardiac performance in fish, yet the cellular mechanisms are largely unknown. Moreover, how heart rate changes throughout an exercise training program remains unclear. This study investigates how training modifies ventricular action potential (AP) properties, which trigger myocyte contraction and influence cardiac excitation-contraction coupling and rhythm. The APs in isolated ventricular cardiomyocytes were measured electrophysiologically, while heart rate in freely swimming fish was monitored using bio-loggers. Fish were divided into control and trained groups. Control fish were kept in standard aquaculture tanks with a water flow rate of 0.3 body lengths per second (bl/s), while trained fish exercised 6 hours, five days a week, for 4 weeks, at 0.9 bl/s. After four weeks, significant electrical remodeling was observed in myocytes of trained fish. The resting membrane potential (RMP) was more depolarized, the action potential duration at 90% repolarization (APD90) was prolonged, and the threshold potential (TP) was significantly more depolarized compared with controls. Bio-logging data showed significant changes in heart rate. During the first three weeks, trained fish had significantly elevated heart rate during exercise sessions. However, by the beginning of week four, the heart rate during exercise of the trained fish was same as heart rate in control fish, and resting heart rate decreased below control levels. The results show that training modulates significantly the AP properties of heart which are connected to changes in heart rate. However, it takes three weeks to see the effects of training on heart rhythm.

MOTION I Metabolism in motion: Mechanics, physiology, and ecology of fish movement

Kinematic modulation for flow exploitation as a foundation for Locomotor energy conservation

Zhang, Yangfan (*yangfan_zhang@fas.harvard.edu*) Oral Presentation
Co-Authors: Lauder, George
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Abstract: Understanding how fish conserve energy while moving through complex flows requires the integration of physiology, biomechanics, and hydrodynamics. Here, we synthesize recent work to propose a unifying perspective: kinematic modulation in flows is fundamental for locomotor energy conservation. Artificial Intelligence-enabled tracking of schooling fish shows that individuals in schools exhibit substantial positional rearrangement and kinematic variability, yet still gain energetic benefits when compared to individual fish swimming alone. A robotic flapping mechanical apparatus demonstrates that fish can modulate their kinematics in response to fluid stimuli from neighbours. These results suggest that fish schools have dynamic kinematic coupling with hydrodynamic structures rather than swimming in fixed formations. Furthermore, we reveal that fish actively exploit velocity gradients to achieve passive thrust and overall force equilibrium, which reduces propulsion effort and metabolic cost towards a resting state while in flows. The variation in metabolic rate is tightly linked to the variation in gait through time. Collectively, these studies suggest that the kinematics of fish can actively interact with flow to harness kinetic energy, which leads to reduced locomotor energy consumption.

MOTION I Metabolism in motion: Mechanics, physiology, and ecology of fish movement

Effects of social environment and energy efficiency on preferred swim speed in a marine generalist fish, pile perch (Phanerodon vacca)

Tiddy, Izzy C.[1] (*isabelle.tiddy@glasgow.ac.uk*) Poster Presentation
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Abstract: Energy efficiency is a key component of movement strategy for many species. In fish, optimal swimming speed (Uopt) is the speed at which the mass-specific energetic cost to move a given distance is minimised. However, additional factors may influence an individual's preferred swimming speed (Upref). Activities requiring consistent sensory inputs, such as food finding, may require slower swimming speeds than Uopt. Further, although the majority of fish display some form of social behaviour, the influence of social interactions on Upref remains unclear. It is unlikely that all fish within a group will have the same Upref, and fish may therefore compromise individual Upref to swim with a conspecific. This study measured the Uopt, Upref and Upref in the presence of a conspecific (Upair) of pile perch, Phanerodon vacca, a non-migratory coastal marine

generalist. Uopt was significantly higher than, and was not correlated with, Upref. Fish therefore chose to swim at speeds below their energetic optimum, possibly because slower swimming allows for greater awareness of surroundings. Mean Upair was significantly lower than the Upref of the faster fish in each pair but did not differ significantly from the Upref of the slower fish. Therefore, faster fish appear to slow their speed to remain with a slower conspecific. Our study suggests that environmental factors, including social surroundings, may be more important than energetic efficiency for determining swim speed in P. vacca. Further studies of fish species from various habitats will be necessary to elucidate the environmental and energetic factors underpinning Upref.

MOTION II Physiology in motion: Linking movement, metabolism, and life history of fishes

Physiology in motion: Linking movement, metabolism, and life history of fishes
Christian Bihun and Erin Stewart

Abstract: Advances in biologging and telemetry technologies are transforming our understanding of fish biology by allowing researchers to track not only where fish go, but how they function in the wild. This session highlights studies that combine fish movement with physiological insights such as metabolic rate, activity patterns, behavioural thermoregulation, and stress responses, using tools such as acoustic or satellite tags, accelerometers, heart rate loggers, and archival sensors. We welcome contributions that take integrative approaches to explore questions related to life history strategies, environmental constraints, bioenergetics, and vulnerability to anthropogenic impacts and climate warming. By bridging the fields of physiology and ecology, this session will showcase the utility of in situ measurements to reveal the inner lives of fishes.

Exploring the ecological relevance of metabolism and bioenergetics for wild fishes: new technological approaches and key questions

Brownscombe, Jacob (*jakebrownscombe@gmail.com*) Oral Presentation

Abstract: Metabolism and bioenergetics are both rich fields of study with fishes, with established relevance in linking environmental conditions with fish productivity and ecological interactions. Most studies in these fields rely upon laboratory-based data, creating challenges in interpreting their relevance for wild fishes facing dynamic environmental conditions. Further, a variety of performance-related metrics have been developed (e.g., linking temperature with aerobic scope or growth), but there remains uncertainty in exactly when, where, and how broadly applicable these metrics are as key ecological indicators. Recent advances in combined experimental approaches and field techniques are enabling studies to generate further insights into the role of metabolic and bioenergetics metrics in wild fishes and aquatic ecosystems. There remain important fundamental questions about how ecological conditions constrain fish population productivity and persistence, which may require a variety of study approaches and data sources that span outside these fields (e.g., population and food web modeling). In this talk I will discuss these challenges, novel study approaches, recent findings, and directions for further research. Energy mobilized via metabolism is an important fundamental currency for animals and natural ecosystems, and hence these studies are some of the most valuable directions for both fundamental and applied fish ecology research.

Quantifying burst swimming in sockeye salmon during their 1100km upstream migration

Birnie-Gauvin, Kim[1] (*kbir@aqua.dtu.dk*) Oral Presentation
Co-Authors: Robinson, Kendra[2]; Byrnes, Evan[3]; Van Wert, Jacey[4]; Venditti, Jeremy[5]; Cooke, Steven[6]; Eliason, Erika[2]; Patterson, David[2]
[1]Technical University of Denmark; [2]Department of Fisheries and Oceans Canada, [3]Stanford University, [4]University of California Santa Barbara, [5]Simon Fraser University, [6]Carleton University,

Abstract: In many fishes, the cost of swimming represents a large component of their energy budget. In particular, energy is an important and limited commodity for upstream migrating adult Pacific salmon that have ceased feeding. Thus, quantifying how these fish partition their energy during their migration to ensure they successfully spawn is of conservation interest. In the Fraser River, some sockeye salmon populations migrate upwards of 1100 km to reach their spawning grounds, and encounter severe hydraulic challenges along the way. Understanding how sockeye salmon partition their finite energy budget into aerobic (sustained) and anaerobic (burst) swimming could help us understand why some fish successfully make it to the spawning grounds while others do not. We tagged sockeye salmon upon their entry in freshwater with archival accelerometer radio tags and recovered 16 tags on the spawning grounds, providing us with the first full migration datasets from which we can quantify burst swimming. We found that the type of bursts (e.g. duration, maxima, peak count, effort) that sockeye salmon perform differs along their migration, likely reflecting river hydromorphology, but that on average, fish only spend 1.23% of their migration bursting. The Fraser Canyon - unsurprisingly - required significantly more bursting than the remainder of their migration, representing 75.3% of the cumulative bursting effort despite representing only 35% of the distance. Our study is the first to quantify burst swimming along 1100km of migration in sockeye salmon.

MOTION II Physiology in motion: Linking movement, metabolism, and life history of fishes

Brook trout in hot (and hypoxic) water

Reeve, Connor (*cjr273@cornell.edu*) Oral Presentation
Cornell University

Abstract: During summer, lake-dwelling brook trout (Salvelinus fontinalis) may experience oxythermal habitat compression due to warming surface waters and progressive deep-water deoxygenation over the stratified period. In regions such as the Adirondacks, this phenomenon is becoming increasingly common due to the combined effects of climate warming and brownification, yet its consequences for this sensitive cold-water species remain poorly understood. To evaluate how compression shapes trout behavior, we implanted fish with temperature and depth sensor equipped acoustic transmitters and released them into lakes spanning a gradient of oxythermal conditions. Additionally, in each lake we deployed vertical arrays of temperature and dissolved oxygen loggers to track temporal changes in habitat suitability. Suitable oxythermal habitat was defined using

thresholds derived from our recent tolerance assays across multiple brook trout strains, which identified a functional upper thermal limit of 21°C and a lower oxygen limit of 3.5 mg L-1. Integrating these criteria with field telemetry, we quantified oxythermal preference and avoidance, and tested how shifts in suitable habitat influenced depth use and foraging-related behaviors. Our findings indicate that the combined effects of warming and deep-water deoxygenation increasingly constrain habitat use and reduce opportunities for effective foraging. As warming and browning continue to erode summer habitat quality, brook trout populations may face increasing limitations on growth, performance, and long-term persistence.

MOTION II Physiology in motion: Linking movement, metabolism, and life history of fishes

The effect of size, age and sexual maturity on acoustic tagging success in an annual, endangered fish species

Steel, Anna[1] (*aesteel@ucdavis.edu*) Oral Presentation
Co-Authors: Gonzales, Sebastian[1]; Chapman, Eric[2]; Cocherell, Dennis[1]; Deng, Zhiqun (Daniel)[3]; Hassrick, Jason[2]; Wilder, Rick[2] and Fangue, Nann[1]
[1]University of California Davis; [2]ICF International, [3]Pacific Northwest National Laboratory

Abstract: Delta smelt, Hypomesus transpacificus, are a federally endangered species endemic to California's Sacramento-San Joaquin Delta. Although acoustic telemetry approaches could provide valuable information to support conservation efforts, implementation has historically been impractical due to the relative size of available acoustic transmitters. Recent advances in transmitter miniaturization by the Pacific Northwest National Laboratory have produced transmitters weighing less than 5% of adult Delta smelt body weight (Shad Tag, 0.05g), bringing renewed interest to the use of acoustic telemetry with this species. Here we present an evaluation of the feasibility of acoustic telemetry tagging methodologies for Delta smelt with these smaller tags. Despite initial evidence-based modifications to surgical methods developed for other small-bodied fishes, post-surgical survival still varied substantially among tagged individuals. Delta smelt exhibit a predominantly annual life-history pattern, and do not reach taggable size until adulthood, making it unavoidable to tag sexually mature individuals. Yet physiological stress and energetic trade-offs associated with gonadal development and reproduction may contribute to variability in tagging success, as measured by survival and ecological performance. Here, we present results from a seven-month study assessing the effects of acoustic tag implantation on survival and swimming performance in a single cohort of Delta smelt. Groups of fish were evaluated as the cohort increased in size and age, and progressed through stages of sexual maturation. Our results inform post-surgical survival assumptions for acoustic telemetry applications in Delta smelt and provide a framework for assessing relationships between ontogenetic state and tagging success in other short-lived or seasonally reproducing fishes.

MOTION II Physiology in motion: Linking movement, metabolism, and life history of fishes

Telemetry tracking and physiological sampling indicate large smolt survival differences at a fish-friendly flood pump

Lingard, Stephanie (*lingard.steph@gmail.com*) Oral Presentation
Co-Authors: Kinnard, Hollis[1]; Hinch, Scott[2]; Eliason, Erika[3] and Lotto, Andrew[1]
[1]University of British Columbia; [2]Faculty of Forestry & Environmental Stewardship;
[3]Fisheries and Oceans Canada

Abstract: Fish are injured in a variety of ways when passing through human infrastructure. The majority of fish passage research has thus far focused on immediate survival impacts resulting from major injuries. However, infrastructure can also have delayed survival impacts through increases in physiological stress, or visually imperceptible injuries. To address questions surrounding the safety of flood-infrastructure for Pacific salmon, we investigated the impact "fish-friendly" infrastructure had on both immediate and delayed indices of survival in juvenile salmon survival over two years (2024 & 2025). We assessed immediate impacts by releasing marked Coho smolts upstream (n=1404) and downstream (n=398) a flood management site in the Fraser Valley, British Columbia. Marked fish were recaptured downstream of the station and assessed for external injuries and indices of anaerobiosis (lactate) and internal injury (hemoglobin). Coho that had passed the infrastructure experienced eye injuries and contusions at a higher rate than those released downstream.In the first year, hemoglobin (77.9 mg/L) was lower in fish passing the facility than in control fish (90.7 mg/L). Delayed impacts were assessed by tracking fish implanted with acoustic transmitters (n=592) to the Fraser River estuary. Survival from release to the estuary varied among years for Coho passing the facility (49% [2024]; 61% [2025]) but was relatively consistent in fish released downstream of the facility (71% [2025]; 73% [2024]). These results suggest even infrastructure marketed as 'fish-friendly' may have negative impacts on salmon and highlight the benefit of applying multiple techniques to assess the safety of infrastructure for Pacific salmon.

MOTION II Physiology in motion: Linking movement, metabolism, and life history of fishes

Evaluating the "fish-friendliness" of flood pumps using Sensor Fish

Kinnard, Hollis[1] (*hollis.kinnard@gmail.com*) Oral Presentation
Co-Authors: Hinch, Scott[2]; Lingard, Stephanie[1] and Straker, Dan[3]
[1]The University of British Columbia; [2]Faculty of Forestry & Environmental Stewardship;
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Abstract: Sensor Fish were created to study smolt passage and survival through large hydropower dams, and have given researchers valuable information about improving fish passage. Despite their usefulness, Sensor Fish have not yet been widely used to study other structures that cause barriers to salmon passage. In the lower Fraser River, flood pumps are abundant, and are thought to cause injury and mortality to out-migrating juvenile salmonids. There has been a recent push to upgrade traditional pump stations to fish friendly pump stations, but there is very little research on how well these friendly

pumps allow fish to pass unharmed. This presentation will explore recent work that uses Sensor Fish to study fish friendliness of flood pumps in the lower Fraser River.

MOTION II Physiology in motion: Linking movement, metabolism, and life history of fishes

Investigating walleye activity with accelerometer-bearing acoustic tags in the Great Lakes

Bihun, Christian[1] (*Christianj.bihun@gmail.com*) Oral Presentation
Co-Authors: Faust, Matt[2]; Dembkowski, Dan[3]; Watkinson, Doug[4]; Johnson, Tim[5]; Brownscombe, Jacob[4] and Raby, Graham[1]
[1]Trent University, [2]Ohio Division of Fish and Wildlife; [3]University of Wisconsin-Stevens Point; [4]Fisheries and Oceans Canada, [5]Ontario Ministry of Natural Resources

Abstract: How do activity patterns influence energy expenditure in fish, and do these patterns vary between sexes and across seasons? Walleye (Sander vitreus), are apex predators of high ecological and economic importance, and offer a unique system to investigate these questions considering that females are ~25% larger than males at a given age (i.e., female-biased sexual size dimorphism). We tracked over 200 walleye from North America's largest populations in Lake Erie, Lake Ontario, Lake Michigan (Green Bay), and Lake Winnipeg over a one-year time frame. Our results demonstrate that walleye activity closely tracks seasonal changes in day length, exhibiting shifting diel rhythms that seem to collapse under ice. We also observed higher female activity during winter months, which may reflect increased foraging to support the energetic demands of gonadal development. Using a calibration model previously established by our lab, we predicted active metabolic rates and daily energy costs from the field-derived measurements of acceleration and temperature. Our study highlights the complexity of integrating sex- and season-specific activity into bioenergetics models and provides a foundation for more refined applications of these tools.

STRESS II Stress in fish I (regulation of HPI axis functioning and response to stress) and II (stress effects on physiology – mechanistic insights)

Stress in fish I (regulation of HPI axis functioning and response to stress) and II (stress effects on physiology – mechanistic insights)

Luis Tort, Matt Vijayan, Nick Bernier and Katie Gilmour

Abstract: The stress response consists of a suite of coordinated physiological and behavioural responses that serve to help fish cope with challenges that threaten their capacity to function. At a neuroendocrine level, these responses reflect the activity of the hypothalamic-pituitary-interrenal (HPI) axis, which is regulated by molecular, epigenetic and physiological mechanisms. In turn, the activity of the HPI axis affects the physiology and behaviour of the animal in ways that depend on the severity and duration of the stressor. “Stress in Fish I: Regulation of HPI axis functioning and response to stress” will attract researchers working on the mechanisms of HPI axis activity and regulation, whereas “Stress in Fish II: Stress effects on physiology – mechanistic insights” will provide a home for research on interactions between stress physiology and environmental factors such as temperature, social interactions or endocrine-disrupting chemicals.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

The effects of PFAS on the stress axis in zebrafish (Danio rerio).

Gorissen, Marnix (*Marnix.Gorissen@ru.nl*) Oral Presentation
Co-Authors: Faught, Erin; Schaaf, Marcel; Smeets, Jolie and Klaren, Peter
Radboud University

Abstract: Both humans and animals are exposed to environmental contaminants that may disrupt hormone signalling essential for physiological homeostasis. One widely present and extraordinarily persistent group of contaminants is per- and polyfluoroalkyl substances (PFAS). However, little is known regarding the effects of these Endocrine Disrupting Chemicals (EDCs) on the development and function of the endocrine stress system.We examined the effects of PFAS on the development and function of the hypothalamus-pituitary-interrenal (HPI) axis, a crucial endocrine axis involved in the release of the stress hormone cortisol. Cortisol plays a key role in the stress response and in maintaining homeostasis. We exposed zebrafish embryos and larvae to individual PFAS, as well as to a mixture of PFAS compounds during critical developmental windows, analogous to the second and third trimester in humans, as well as post-natal exposure. The selected PFAS, viz., PFOS, PFOA, PFHxS, and PFHpA, are highly prevalent in the blood of the Dutch population, reflecting significant human exposure. Our results show that postnatal exposure to PFHxS, both individually and in a mixture, resulted in sustained cortisol levels following stress. Moreover, glucocorticoid receptor (GR) signalling remained intact. Future

work will determine how sustained cortisol levels impact stress-related behaviour and will further elucidate the molecular mechanisms of PFAS signalling. Overall, our findings indicate that exposure to these so-called 'forever chemicals' can fundamentally alter stress physiology, highlighting the urgent need for the remediation.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Early life Glucocorticoid Overexposure Primes Cell-Type Specific Stress Hypersensitivity in Adult Zebrafish

Choi, Min-Kyeung (*m.choi@exeter.ac.uk*) Oral Presentation
Co-Authors: Ryu, Soojin and Tochwin, Anna
University of Exeter

Abstract: Early-life exposure to elevated glucocorticoids (GCs) is associated with increased vulnerability to stress later in life. Such effects are often not evident under basal conditions but becomes apparent under stress conditions. The hypothalamic cell populations that encode this hidden stress sensitivity, and the mechanisms underlying its long-term persistence, remain poorly defined. Here, we asked which neuroendocrine cells in the adult zebrafish preoptic area (POA) (a central regulator of the Hypothalamic-Pituitary-Interrenal gland axis) respond to acute stress and whether a history of early-life GC overexposure (eGC-OE) alters their responses. We used a zebrafish "double-hit" paradigm combining optogenetic developmental GC elevation with an acute predatory-like stimulus in adulthood (looming dots, LD). Following LD-exposure, eGC-OE fish showed exaggerated endocrine responses and altered brain gene expression compared to controls. To resolve these effects at cellular resolution in the POA, we performed single-nucleus Multiome profiling of the adult POA. Acute stress selectively activated specific subsets of neuroendocrine subtypes, including a glutamatergic somatostatin (sst1.1+) population and two arginine vasopressin (avp+) clusters. Although eGC-OE had little effect on basal gene expression, it markedly amplified stress-evoked transcriptional responses in these subtypes. Gene regulatory network analyses suggest increased convergence of GR-, CREB-, and MAPK-linked signaling onto immediate-early gene networks associated with neural activation and neuropeptide release mechanisms. Pharmacological perturbation of identified targets reduced the exaggerated cortisol stress response in eGC-OE fish supporting a functional link between altered transcriptional amplification and endocrine hypersensitivity in eGC-OE fish. Together, these findings identify discrete hypothalamic populations that encode latent stress sensitivity programmed by early-life GC exposure.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Interactive effects of thermal and social stress on developmental stress physiology in zebrafish

Vámos, Sofia (*sofia.duartevamos@ucdconnect.ie*) Oral Presentation
Co-Authors: Wollenberg Valero, Katharina
University College Dublin

Abstract: Early-life stress can shape the regulation of stress physiology, yet how environmental and socially mediated stressors influence behavioural outputs during early development remains poorly understood. In fish, behavioural reactivity provides a functional readout of hypothalamic-pituitary-interrenal (HPI) axis activity and its modulation by external challenges. We investigated how thermal stress and stress-derived chemical cues alter stress-related behaviour in zebrafish (Danio rerio) embryos and larvae. Using a high-throughput photomotor response (PMR) assay at 1 day post-fertilization, we quantified locomotor reactivity following acute light stimuli under control, thermal stress, and stress-metabolite conditions across multiple rearing densities. Embryos exposed to elevated temperature or to metabolites released by heat-stressed conspecifics showed significant shifts in response magnitude and recovery dynamics, indicating altered neural stress sensitivity prior to full maturation of the HPI axis. Rearing density further modulated these effects, demonstrating that social context interacts with environmental stress to shape early behavioural phenotypes. Follow-up assays at 4 days post-fertilization showed that early stress exposure alters subsequent startle responsiveness and stress-related behavioural patterns, suggesting that transient embryonic stress can recalibrate later behavioural reactivity. Together, these findings show that thermal stress is not only directly experienced but can be socially transmitted during development, with measurable consequences for stress-related behaviour. By integrating environmental, social, and developmental aspects, we aim to better understand how early-life conditions shape behavioural outputs linked to HPI-axis function under climate-relevant thermal stress.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Nucleobindin-derived peptides cause stress in male zebrafish (Danio rerio)

Sethi, Paalki (*rsd431@usask.ca*) Oral Presentation
Co-Authors: Unniappan, Suraj; Rachamalla, Mahesh and Niyogi, Som
University of Saskatchewan

Abstract: Rationale: Stress is a fundamental physiological response comprised of complex neuroendocrine and behavioural interactions. Prolonged stress leads to anxiety, depression, and metabolic disorders in animals. In zebrafish, the hypothalamus-pituitary-interrenal (HPI) axis hormones are crucial for stress regulation. Emerging evidence indicates additional molecular players may modulate stress. Nucleobindin (NUCB)-derived, feeding and metabolism-regulating peptides, nesfatin-1 (derived from NUCB2) and nesfatin-1-like-peptide (NLP; processed from NUCB1) are expressed in key stress-regulatory brain regions. Stress and metabolism are tightly interlinked. We hypothesizedthat nesfatin-1 and NLP cause stress-related behaviour and modulate HPI

axis components in zebrafish. Methods: Nesfatin-1 and NLP (100ng/g B.W.), cortisol (positive control; 100ng/g B.W.), and saline (control) were intraperitoneally injected into male zebrafish. Behavioral assays (novel tank test, social preference and shoaling behavior) were performed 10 minutes post-injection. Injections were repeated after two weeks, and HPI tissues were collected after 1 hour for gene analysis. Results: Acute administration of nesfatin-1 or NLP increased locomotor activity, exploratory response, increased social approach and fewer excursion events (1-way ANOVA, $19.76<F_{4,85}<215.6$, $P<0.0001$). RT-qPCR revealed significant changes in HPI axis mRNAs. Specifically, an increase in brain crf (corticotropin-releasing factor) mRNA after nesfatin-1 treatment and interrenal gr (glucocorticoid receptor) mRNA post-NLP treatment was observed. Whereas, a decrease in brain crf-r1 and interrenal 11β-hydroxylase (steroidogenesis) in both nesfatin-1 and NLP treatment groups (1-way ANOVA, $3.73<F_{4,20}<9.06$, $0.0008<P<0.006$) was found. Conclusion: Nesfatin-1 and NLP cause stress-related behaviour and modulate key endocrine components of the HPI axis in male zebrafish. These peptides have the potential for further exploration as stress biomarkers.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Brain drain: applying dynamic energy budget theory to the regulation of adult neurogenesis

Alderman, Sarah (*alderman@uoguelph.ca*) Oral Presentation
Co-Authors: Aitken, Lindsey
University of Guelph

Abstract: The Dynamic Energy Budget Theory is a helpful framework for predicting trade-offs in expensive tissues during periods of limited resources. The brain is the most expensive tissue in vertebrates, and fishes invest heavily in brain growth via lifelong neurogenesis. Chronic stress inhibits neurogenesis via cortisol mediated inhibition of brain cell proliferation, which may serve to temper investment in the brain during high allostatic load; however, this has not been explicitly tested. We hypothesized that individual rates of neurogenesis are modified to offset long-term energy deficits. An acute (3-day) and chronic (28-day) negative energy balance were imposed on zebrafish in a factorial experiment consisting of two temperatures (28°C, 33°C) and two feeding regimes (fed, fasted/caloric restriction) to increase energy expenditure and limit energy intake, respectively. Reduced condition factor in zebrafish held at 33°C with restricted feeding confirmed a negative energy balance at both timepoints. Cell proliferation in the telencephalon was quantified using mitotic cell labeling (EdU). The number of EdU+ cells in the telencephalon did not differ between experimental groups in either the acute or chronic experiments suggesting organismal energy status did not influence the rate of brain cell proliferation. Instead, we provide evidence of trade-offs with other expensive tissues (e.g., gut mass) and processes (e.g., female reproduction) during a chronic negative energy balance. These findings implicate neurogenesis as a critical process in zebrafish that is prioritized even as investment in somatic maintenance, gut mass, and reproduction are reduced to accommodate a negative energy balance.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Cortisol alters the hypothalamic lipidome in rainbow trout

Antomagesh, Femila[1] (*femilarani.antomages@ucalgary.ca*) Oral Presentation
Co-Authors: Blanco, A.M.[2]; Buzatto, A.Z.[1]; Soengas, J.L.[2]; and Vijayan, M.M.[1]
[1]University of Calgary; [2]Universidade de Vigo

Abstract: The hypothalamus centrally regulates nutrient intake through two opposing neuronal clusters: the orexigenic agouti-related peptide (AgRP)/neuropeptide Y (NPY) and the anorexigenic cocaine- and amphetamine-regulated transcript (CART)/proopiomelanocortin (POMC). However, the stress regulation of these neuronal clusters in fish are unclear. We previously reported that cortisol-induced feeding inhibition in rainbow trout (Oncorhynchus mykiss) is associated with altered hypothalamic metabolome and enhanced gene transcripts, and activities of enzymes involved in fatty acid biosynthesis. Consequently, we hypothesized that cortisol inhibits feeding by promoting an energy rich hypothalamic environment by increasing the lipid availability. To test this, the trout were given a slow-release cortisol implant for 3 days, followed by intracerebroventricular injection of firsocostat, an inhibitor of fatty acid biosynthesis. Hypothalamus was collected 1 h post-injection for untargeted lipidomics. We identified 432 lipid species across 21 classes. Chronic cortisol selectively increased phosphatidylserine (PS) and phosphatidic acid (PA) while decreasing triglycerides. The PS and PA are essential for neuronal membrane remodeling, exo/endocytosis, and synaptic signaling. Triglycerides are key fuels during elevated synaptic activity. At the lipid species level, an enhanced PS 18:2_22:4 and PA 18:2_22:4 abundance, implicates linoleic acid-enriched phospholipid remodeling. Overall, our data provide the first lipidomic characterization of the trout hypothalamus and the changes under chronic stress and/or cortisol stimulation. Future studies will characterize whether some of these lipid species may be acting as key signaling molecules contributing to the feeding cessation during stress in trout. Study was supported by the Natural Sciences and Engineering Research Council of Canada Discovery grant to Dr. Vijayan.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Investigating cortisol as a mediator of acute thermal tolerance in the Atlantic killifish (*Fundulus heteroclitus*)

Flatt, Emma (*eeflatt@zoology.ubc.ca*) Oral Presentation
Co-Authors: Eisenberg, Rachel and Schulte, Patricia
The University of Brirish Columbia

Abstract: The HPI-axis is implicated as a potential mediator of thermal tolerance in fish, as shifts in cortisol, are associated with improvements or reductions in the critical thermal maximum (CTmax). The Atlantic killifish (Fundulus heteroclitus) has two geographically distinct subspecies that differ in CTmax and stress reactivity. Given this, we hypothesized

that the latitudinal thermal range of these subspecies determines the activation threshold of the cortisol response altering stress phenotypes and determining acute thermal limits. We tested this hypothesis by inhibiting cortisol synthesis using metyrapone and assessing the extent of cortisol release during a CTmax trial in both metyrapone treated and untreated southern and northern killifish to determine if subspecies differences in CTmax can be ablated by metyrapone administration. When cortisol was inhibited, CTmax was significantly decreased for northern killifish but subspecies differences were maintained with an essentially equivalent reduction in CTmax. The agitation temperature of these fish, noted as escape behaviours displayed during the trials such as burst swimming or jumping, was also significantly reduced in both subspecies when cortisol was inhibited. These results provide further support for cortisol-mediated effects on acute thermal limits and suggest a potential role for cortisol in thermal reception. On going analysis of waterborne cortisol samples taken at select timepoints during the thermal ramp and brain, head kidney, liver and gill gene expression will help further elucidate the relationship between cortisol levels and CTmax.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Cellular thermal profiles in the mucus of wild bull trout

Lazaro-Côté, Analisa[1] (*a.lazarocote@gmail.com*) Oral Presentation
Co-Authors: Lothian, Angus[2]; Kissinger, Benjamin[3]; Glowa, Sarah[4]; Mochnacz, Neil[4] and Jeffries, Kenneth[1]
[1] University of Manitoba; [2]University of Calgary, [3]fRI Research, [4]Fisheries and Oceans Canada

Abstract: With water temperatures increasing worldwide, many aquatic ectotherms are facing more frequent and severe thermal conditions pushing them to their physiological limits. Transcriptional profiling can be used for early detection of sublethal thermal stress because shifts at the cellular level are identified earlier than harmful organismal-level impacts. However, for many sensitive species lethal or invasive sampling is prohibited, limiting our knowledge of their physiology and adaptive potential with respect to warming temperatures. In lab-based studies, we found that epidermal mucus mirrors thermal thresholds in commonly sampled tissues, indicating that mucus transcriptional profiling offers a systemic and non-invasive approach to assess the physiological status of individuals. Therefore, we collected mucus swabs from wild bull trout, a species legally protected under Canada's Species at Risk Act. We targeted individuals from their northern (Nahʔą Dehé, Northwest Territories) and central (Kakwa, Alberta) Canadian range extents. These populations differ in the thermal regimes and selection pressures they experience, including the severity of anthropogenic impacts. Our objective was to determine the influence of thermal regimes and anthropogenic disturbances on transcriptional profiles in mucus. We measured transcript abundance using a high-throughput transcriptional profiling chip and applied a random forest classifier to identify individuals at high risk of thermal stress. This enabled comparison of temperature thresholds for cellular stress activation between populations. Our study highlights the use of transcriptional breakpoints

to determine when temperatures begin to be harmful to fish, allowing resource managers to intervene before population declines are observed.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Transcriptional profiling provides insights into how fasting affects the thermal tolerance and physiological response to acute thermal stress in lake sturgeon

Bernier, Nick[1] (*nbernier@uoguelph.ca*) Oral Presentation
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Abstract: Lake sturgeon (Acipenser fulvescens) populations across their North American native range remain classified as threatened despite decades of recovery efforts. Since periods of rapid summer growth at optimal temperatures are vital for juvenile recruitment, both thermal stress and reduced food availability may pose significant challenges to the recovery and survival of this species. In this study, to characterize the potential threats posed by decreased energy reserves to the physiological response to acute thermal stress in lake sturgeon, we examined how fasting for periods of 5 and 10 weeks affects the endocrine stress response and the transcriptomic response to a CTmax test. Relative to fed fish, prolonged fasting enhanced upper thermal tolerance and reduced the cortisol stress response to acute thermal stress. Targeted liver and gill transcriptional profiling indicated that fasting shifts energy use from glucose to fat metabolism, decreases glycolytic capacity and oxidative phosphorylation, and downregulates the endocrine growth axis. Conversely, compared with fed fish, fasted fish following acute thermal stress exhibited gene expression profiles indicative of reduced antioxidant capacity and increased cellular stress response. Overall, these results provide mechanistic insights into the potential trade-offs involved in physiological responses to decreased energy reserves and acute thermal stress in lake sturgeon, as well as their ability to cope with these stressors.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

miR-140-5p as a biomarker of thermal challenges in the European seabass (Dicentrarchus labrax)

Sniecinski, Elisa[1,2] (*elisa.sniecinski@ifremer.fr*) Oral Presentation
Co-Authors: Bégout, Marie-Laure[1,2]; Blanc, Marie-Odile[1,2]; Dutto, Gilbert[1,2]; Maire, Anthony[3]; McKenzie, David J.[1,4] and Geffroy, Benjamin[1,2]
[1]MARBEC, [2]IFREMER, [3]Électricité de France, [4]Centre National de la Recherche Scientifique

Abstract: MicroRNAs (miRNAs) are small non-coding RNAs, typically of 20-22 nucleotides, that play a critical role in the epigenetic regulation of numerous biological processes. Under specific environmental conditions, miRNAs regulate the expression of target genes by binding to the three-prime untranslated region (3'UTR) of messenger RNAs, thereby inhibiting or promoting their translation into proteins. Recent studies suggest that these epigenetic mechanisms may be crucial for fishes to cope with environmental stressors. We

previously demonstrated that miR-140-5p is differentially expressed following short- and long-term thermal challenges. Hence, here we investigated the role of this miRNA in the regulation of potential target genes in response to an acute increase in temperature. Our model is the European seabass (Dicentrarchus labrax), an important species for aquaculture and fisheries. Fish were injected intraperitoneally with either a miR-140-5p mimic, a miR-140-5p inhibitor, a saline solution, or were left uninjected as treatment controls. Fish were then subjected either to an acute temperature increase (+6°C over 6 hours) or maintained at under normal rearing conditions at 21 °C, as temperature controls. After 6 hours, samples of plasma and interrenal gland were collected. RT-qPCR confirmed that the injection treatments successfully modified miR-140-5p expression in the interrenal glands. RNA sequencing to evaluate potential target gene expression revealed differential expression of potential miR-140 target genes involved in cortisol regulation. These results suggest that miR-140-5p may act as an epigenetic marker involved in the response to thermal challenges in fishes.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Using transcriptomics to explore variation in cold stress physiology and resilience of gizzard shad (Dorosoma cepedianum) to cold snaps

Waraniak, Justin[1] (*waraniak@illinois.edu*) Oral Presentation
Co-Authors: Suski, Cory[1]; Esbaugh, Andrew[2]
[1] University of Illinois Urbana-Champaign; [2] University of Texas Marine Science Institute

University of Illinois Urbana-Champaign; University of Texas Marine Science Institute

Abstract: While much research has rightly concentrated on effects of warmer temperatures on the stress physiology of fishes, extreme cold events still pose a risk for many species, and cold physiology is relatively understudied.Clupeid fishes are known to be susceptible to colder temperatures, and cold snaps have been linked to die-offs of gizzard shad (Dorosoma cepedianum) and other clupeid fishes. Transcriptomics have emerged as a method to monitor different types of stress in wild populations and possibly attribute the mortality to cold stress or other sources. In this study, we aim to develop biomarkers of cold stress for gizzard shad. We collected shadon two separate occasions from Lake Erie (N=32; collected at 20 ºC) and the Illinois River (N=32; 14º C) and subjected them to acute cold stress trials. Fish were split into treatment and control groups, and treatment groups were first subjected to a mild cold stress (12 ºC for Lake Erie, 9 ºC for the Illinois River) for 1 hour before half (N=8) had tissues sampled for transcriptomic analysis. The remaining fish (N=8) were then chilled to test their critical thermal minima (CTmin) and sampled. Three tissues: gill filaments, muscle, and liver, were collected from each fish. RNA expression data was generated from these samples and analyzed using differential expression analyses to identify significantly up/down-regulated genes. We also found differenced in the CTminbetween fish collected from Lake Erie (6.8 ± 2.1 ºC, mean ± sd) and the Illinois River (3.9 ± 0.6).

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Leptin function in hypoxia and its implications in the adaptive stress response in tilapia

Borski, Russell[1] (*russell_borski@ncsu.edu*) Oral Presentation
Co-Authors: Lee, Jr., William[1]; Reynolds, Hannah[1]; Bernier, Nick[2]; Mankiewicz, Jamie[1]
[1]North Carolina State University; [2]University of Guelph

Abstract: Leptin is a potent anorexigenic hormone that stimulates energy expenditure in mammals, but its function in the control of metabolism in fish remains poorly understood. Our previous work in tilapia (Oreochromis spp) shows the hormone suppresses appetite, increases in response to various stressors, is modulated by epinephrine and cortisol and may mobilize carbohydrate stores. Leptin also stimulates glycolysis and lactate production under normoxic conditionsin vitro, indicating the hormone directly augments the metabolic pathway common to anaerobic and aerobic respiration. We hypothesize that the hormone may play a role in shifting metabolic processes in response to hypoxic stress. To this end, we evaluated gene networks and the leptin response in the liver, the main source of leptin production, during a 2 and 6 h acute exposure to hypoxia in tilapia (Oreochromis niloticus). We found that blood glucose and lactate increase along with hepaticlepaand muscle and liver leptin receptor (lepr). These responses were associated with alterations in hepatic gene networks (liver transcriptome) associated with hypoxia, including key elements of the hif1a signaling (egln), glucose metabolism, glycolysis, insulin and insulin signaling, erythropoietin (epo), vascular endothelial growth factor (neovascularization), amongst other pathways, many of which correspond with the known actions of leptin, including its function in inducing hypoxic responsive genes. Collectively, these results suggest leptin may facilitate hypoxic adaptation by potentiating a shift towards anaerobic metabolism through increased glycolysis, promoting angiogenesis, and regulation of other physiological processes.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Stress endpoints in hatchery-raised striped mullet, Mugil cephalus, in response to salinity challenge.

Merlo, Reilly (*Merlor@hawaii.edu*) Oral Presentation
University of Hawaii

Abstract: The euryhaline striped mullet, Mugil cephalus, is a culturally important food fish globally, including in Hawai‘i. As a catadromous species, young mullet transition from seawater to estuary environments which have naturally fluctuating water parameters including salinity. To improve our understanding of how the mullet adapt to variable environmental conditions, we tested the response of hatchery-reared mullet to low salinity acclimation and short term rearing in an inland facility. Specifically, we tested the effects of salinity on survival, condition factor, and stress endpoints including cortisol and lipid peroxidation over 9 weeks. Young hatchery-reared mullet (3.89 ± 0.24 g) were received at an inland research facility and placed in artificial seawater (32 ppt). Fish were then

transitioned into four treatment groups: seawater control, brackish water (16 ppt), fresh water (4 ppt), and a fluctuating salinity regime that varied daily, between 12 and 26 ppt, to simulate an estuarine environment. At the onset of the trial, ectoparasites were detected in all tanks and treated. Interestingly, the survival of the mullet with ectoparasites present decreased at lower salinities. Condition factor was the lowest when parasites were first detected, then increased during parasite treatment. Serum cortisol was lowest at time 0, then generally rose in all salinity groups thereafter. By 9 weeks, cortisol was lowest in brackish water fish. Salinity had no effect on serum glucose concentrations. These results suggest that both salinity and stress from ectoparasite infection elicit adaptive physiological responses in mullet.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Sub-lethal impacts of sea lamprey (Petromyzon marinus) parasitism on the stress responses and energy reserves of rainbow trout (Oncorhynchus mykiss).

Czuba, Karolina[1] (*karolinaczuba1@gmail.com*) Oral Presentation
Co-Authors: Quijada-Rodriguez, Alex[1]; Borowiec, Brittney[2]; Alsop, Derek[1] and Wilkie, Mike[1]
[1]Wilfrid Laurier University, [2]McMaster University

Abstract: Sea lamprey (Petromyzon marinus) are a jawless fish native to the Atlantic Ocean that invaded the Laurentian Great Lakes in the early 1900s. Predation of native fishes by these blood-feeding parasitic fish contributed to major declines in native fisheries. Capable of killing up to 21kg of fish over their lifetime, sea lamprey remain a significant threat, particularly to fishes of cultural importance to Indigenous communities, as well as commercial and sport fisheries. Although ecological impacts of sea lamprey are well recognized, it remains unclear: (i) whether sea lamprey parasitism directly causes host mortality, or (ii) whether parasitism causes sub-lethal physiological effects, making hosts more susceptible to environmental stressors. To examine these questions, we exposed rainbow trout (Oncorhynchus mykiss) to juvenile sea lamprey, allowing volitional feeding for up to three weeks. Following exposure, plasma and tissue samples were collected to assess metabolic and biochemical indicators of stress and energy status. Measurements of body mass, length and inspection of intestinal contents confirmed that lamprey feeding occurred. Variation in feeding severity upon trout led us to develop a wound scoring system to classify wounds as moderate or severe based on visible characteristics. We found that parasitism led to depletion of liver glycogen and reduced plasma glucose. Liver size and water content increased, whereas muscle water content remained unchanged. Hematocrit, hemoglobin, and mean cell hemoglobin were unaffected. Despite continued feeding, this pattern reflects elevated metabolic demand during prolonged parasitic exposure. We conclude that sea lamprey parasitism has sublethal effects through sustained metabolic costs.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Social history and stress adaptation: oxidative stress and mitochondrial adaptation in cichlid fish

Dijkstra, Peter (*dijks1p@cmich.edu*) Oral Presentation
Central Michigan University

Abstract: Competition for mates and resources is a major driver of animal behavior, physiology, and health, with consequences for evolutionary fitness. This talk explores how the African cichlid Astatotilapia burtoni copes with the metabolic demands of social competition and reproduction, focusing on oxidative stress and mitochondrial adaptation. In this species, dominant, territorial males engage in frequent aggression and invest heavily in reproduction, while subordinate males are socially suppressed and reproductively inactive. I will discuss how these divergent social trajectories shape the links between gonadal development, antioxidant defenses, and mitochondrial performance. Additionally, I will present experiments using acute hypoxia as a stressor to examine how social history and reproductive investment influence resilience to oxidative challenges, with emphasis on mitochondrial function and oxidative stress in the brain. Together, these results highlight the interplay between social environment, energy metabolism, and stress physiology, revealing how social experience modulates bioenergetic capacity and oxidative stress management.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Modulation of stress responses through the diet: a natural additive to improve gilthead seabream (Sparus aurata) resilience during transport

Cartan, Sara[1] (*sara.cartan@uca.es*) Oral Presentation
Co-Authors: Mancera Romero[1], Juan Miguel[1]; Jerez-Cepa, Ismael[1]; Betancor, Mónica[2] and Astola, Antonio
[1]University of Cádiz, [2]University of Stirling

Abstract: Live fish transport and high-stocking density are common aquaculture practices that trigger stress responses, altering different biological processes in farmed fish. The search for new strategies to attenuate the stress response is essential to ensure productivity. Dietary supplements with sedative properties are a potential solution to reduce the negative effects of stress. Our study assesses the effectiveness of a natural additive with sedative properties (TecnoGMP) on gilthead seabream (Sparus aurata) juveniles. Fish were fed the supplemented diet for three months under low- and high-stocking density conditions before undergoing a 3-hour transport simulation and a subsequent 24-hour recovery. The transport induced the activation of the hypothalamic-pituitary-interrenal axis, confirmed by increased plasma cortisol levels. This activation and the subsequent recovery dynamic were affected by the previous chronic stress induced by the high-stocking density. The use of the phytogenic in the aquafeed modulated the stress response induced by both high density and transport simulation. Hierarchical clustering heatmap revealed a clear separation between sampling points; however, within the

recovery period, the groups were separated primarily by diet, showing that the use of the phytogenic additive in the aquafeed could improve the recovery period after stress. Overall, these findings demonstrate the potential of TecnoGMP to mitigate transport-induced stress and highlight the importance of the previous rearing conditions when facing stressful situations.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

The air breathing physiology and temperature and hypoxia tolerance of the oriental weatherloach, (Misgurnus anguillicautatus)

Burbules, Katherine (*Kburb95@gmail.com*) Oral Presentation

Abstract: Air breathing has evolved in numerous fish species as an adaptation to extreme conditions. As the global climate changes, these species have an inherent advantage over fishes relegated to aquatic respiration alone. The oriental weather loach, Misgurnus anguilicaudatus, has a modified hindgut that facilitates oxygen uptake through the intestinal walls of swallowed air bubbles. We hypothesized that air breathing would facilitate higher tolerance to elevated temperatures and/or hypoxia. To investigate this, we employed a closed system that measured combined aquatic and atmospheric oxygen consumption ($\dot{M}O_2$) as well as gill and air breathing ventilation frequency in unrestrained, undisturbed loaches. Additionally, loaches were examined during an induced activity experiment to examine their ventilatory response to stress. When exposed to increasing degrees of hypoxia (21 kPa to 5 kPa) at 23◦C, air breathing frequency had a non-linear relationship with hypoxia. Additionally, gill ventilation frequency initially decreased then peaked as hypoxia intensified. Average $\dot{M}O_2$, critical partial pressures for regulating oxygen (PCrit), as well as the kPa at which loss of equilibrium occurred increased as temperature increased, as expected. However, open respirometry, induced activity, and ventilatory changes in response to hypoxia had unexpected results. This study provides insight into the respiratory response of tropical air-breathing fish under conditions found in natural populations. Moreover, these results contribute to understanding the implications of severe and permanent climate change on those populations in the future should global temperature trends continue.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Activation of the hypothalamic-pituitary-interrenal axis in fish embryos

Manzon, Rachel (*rachelrem@outlook.com*) Poster Presentation
Co-Authors: Manzon, Richard G. and Sander, Mia
University of Regina

Abstract: Embryos lack mobility and have limited capacity for physiological adaptation; therefore, minor disruptions during rapid growth and change often lead to mortality and long-term effects post-hatch. Global climate change and anthropogenic activities are altering near-shore fish spawning grounds. Given their vulnerabilities, a better

understanding of the physiological adaptive capacity of embryos is paramount. The consensus has been that the hypothalamic-pituitary-interrenal (HPI) axis is not developed in fish embryos, and thus not responsive to stressors until 1-2 weeks post-hatch. However, work by our research group and collaborators has shown that hypoxia stress activates both the HPI-axis and cortisol production in Lake Whitefish during critical developmental windows. Here we expand on previous work to include Walleye (Sander vitreus), an important commercial and sport fish with cultural significance. We found that crhb, pomca, and cyp11a1 transcripts increased early in embryogenesis, remained elevated until the last third of embryogenesis, then decreased to variable extents. Data on whole-body cortisol are pending. Importantly, HPI-axis transcripts were detected as early as two days post-fertilization following handling and hypoxia stress. The more notable increases in crhb, pomca, and cyp11a1 transcripts occurred at 2, 5 (or 9) and 11 days post-fertilization, roughly corresponding to the stages when HPI-axis and cortisol synthesis are activated in Lake Whitefish.These data emphasize the importance of investigating HPI-axis activation during fish embryogenesis and its effect on development.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Effects of prolonged heatwaves on facultative air-breathing, metabolism, and hypoxia tolerance in adult Corydoras aeneus

Rippamonti, Jessica (*Jessica.Rippamonti@unt.edu*) Poster Presentation
Co-Authors: Berggren, Benjamin; Brown, Imogen; Alvarez, Infinity and Dzialowski, Edward
University of North Texas

Abstract: Climate warming is increasing the incidence of global extreme weather events.Heatwaves present added stress to aquatic ectotherms living in shallow, slow-moving rivers and streams, and force animals to adapt to a harsher environment.Here, we investigated how a facultative air-breathing fish, Corydoras aeneus, handled a sustained heatwave, and if air breathing helped counteract the physiological changes that occur in a limited oxygen environment.Adult Corydoras were maintained at either 25oC or an elevated temperature of 32oC for three weeks.We measured the fish's ability to acclimate to prolonged thermal stress after 0, 1.5, and 3 weeks exposure to the heatwave.Frequency of air breathing (fab) and gill ventilation rate (fgv) was assessed at all time points using video recording and flow-through respirometry at 100% and 20% oxygen air saturation.After 3 weeks exposure, we assessed critical thermal maximum (CTmax) and collected posterior gut (air-breathing organ) and gills for morphological assessment.Mitochondria function was measured in cardiac myocytes and gills using high-resolution respirometry to assess changes in cellular metabolic function.While an air-breathing adaptation may be beneficial to handle acute heat stress, the results of this study will better inform our understanding of the ability of air-breathing fish to tolerate widespread environmental effects of climate warming.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Effects of a dietary antioxidant additive on the tissue-specific oxidative status of European seabass (*Dicentrarchus labrax*) reared at different stocking densities.

Blázquez-Durán, Alejandro[1] Poster Presentation
Co-Authors: Mancera Romero, Juan Miguel[1], Martínez-Antequera, Francisca Purificación[2] and Jerez-Cepa, Ismael[1]
[1]University of Cádiz, [2] University of Almería

Abstract: Although high stocking densities reduce aquaculture production costs and increase profits, they often trigger chronic stress, which negatively affects fish metabolism and growth. Consequently, dietary nutraceuticals are increasingly being used to mitigate stress induced by inappropriate stocking densities and improve animal welfare. This study evaluated the efficacy of a phytogenic antioxidant additive (AquaNeox) in European seabass (Dicentrarchus labrax) juveniles reared under different stocking densities. Juvenile fish (105 ± 30 g initial weight) were distributed into four triplicate groups, combining two diets (control or diet supplemented with nutraceutical compound at 0.05%) and two initial stocking densities (low stocking density, LSD, 5 kg/m3; and high stocking density, HSD, 37 kg/m3). After two months of feeding to apparent satiation, fish were sampled and liver and gill were taken to assess the oxidative status. Regarding oxidative status, hepatic tissue showed no significant alterations in response to either stocking density or additive, exhibiting only non-significant fluctuations in antioxidant enzymes. In contrast, gills exhibited a distinct modulation. Although not significant, there was a general trend towards increased basal superoxide dismutase (SOD) and catalase (CAT) activities in fish receiving supplemented diet across both densities. Notably, under HSD condition, fish fed the additive exhibited a significant enhancement in gill glutathione peroxidase (GPx) activity compared to fish at LSD condition. In conclusion, although there was no hepatic effect, phytogenic supplementation primed basal gill antioxidant defences, facilitating a reactive response against high-density oxidative challenges.

STRESS II Stress in fish I (regulation of HPI) and II (stress effects on physiology)

Modulation of the antioxidant enzyme capabilities of European sea bass (*Dicentrarchus labrax*) additive under high stocking density conditions through of a dietary relaxing phytogenic

Blázquez-Durán, Alejandro[1] Poster Presentation
Co-Authors: Mancera Romero, Juan Miguel[1], Martínez-Antequera, Francisca Purificación[2] and Jerez-Cepa, Ismael[1]
[1]University of Cádiz, [2] University of Almería

Abstract: Although high stocking densities reduce aquaculture production costs and increase profits, they often trigger chronic stress, which negatively affects fish metabolism and growth. Consequently, dietary nutraceuticals are increasingly being used to mitigate stress induced by inappropriate stocking densities and improve animal welfare. This study evaluated the efficacy of a phytogenic antioxidant additive (AquaNeox) in European seabass (Dicentrarchus labrax) juveniles reared under different stocking densities. Juvenile

fish (105 ± 30 g initial weight) were distributed into four triplicate groups, combining two diets (control or diet supplemented with nutraceutical compound at 0.05%) and two initial stocking densities (low stocking density, LSD, 5 kg/m3; and high stocking density, HSD, 37 kg/m3). After two months of feeding to apparent satiation, fish were sampled and liver and gill were taken to assess the oxidative status. Regarding oxidative status, hepatic tissue showed no significant alterations in response to either stocking density or additive, exhibiting only non-significant fluctuations in antioxidant enzymes. In contrast, gills exhibited a distinct modulation. Although not significant, there was a general trend towards increased basal superoxide dismutase (SOD) and catalase (CAT) activities in fish receiving supplemented diet across both densities. Notably, under HSD condition, fish fed the additive exhibited a significant enhancement in gill glutathione peroxidase (GPx) activity compared to fish at LSD condition. In conclusion, although there was no hepatic effect, phytogenic supplementation primed basal gill antioxidant defences, facilitating a reactive response against high-density oxidative challenges.

NEURO Fish integrative neurobiology in changing environments

Fish integrative neurobiology in changing environments

Martin Tresguerres, Mar Huertas and Trevor Hamilton

Abstract: The nervous system serves as the sensory interface between an animal and its environment, enabling it to regulate bodily functions, locate food and suitable habitats, interact with mates, and avoid predators. The underlying neurobiological mechanisms have evolved and adapted to naturally fluctuating environments. However, human activities are now introducing rapid environmental changes that may profoundly impact many of these processes. Importantly, neurons do not operate in isolation in real-world conditions. Instead, they function within internal microenvironments maintained by various other cells (e.g., glial cells), tissues, and organs (e.g., blood-brain barrier, gills, lungs, kidneys) that ultimately shape behavior. Therefore, understanding the vulnerability and resilience of nervous system function requires a truly integrative approach. This includes multiple fields of comparative physiology—such as neurobiology, behavioral neuroscience, endocrinology, acid-base and ionic regulation, thermal physiology, and metabolism—as well as insights from ecology, genomics, and environmental science. This session will bring together transdisciplinary scientists from these and other diverse areas to discuss the physiological mechanisms that enable animals to maintain neural function during environmental fluctuations—both natural and anthropogenic. Participants will also explore the broader evolutionary, ecological, and conservation/restoration implications of these mechanisms.

NEURO Fish integrative neurobiology in changing environments

Resilient Brains: Behavioral and Physiological Homeostasis in Highly Variable Environments

Hofmann, Hans (*hans@utexas.edu*) Oral Presentation
The University of Texas

Abstract: Anthropogenic climate change increases environmental variability (e.g., prolonged droughts, more severe weather events), posing potent challenges to homeostasis, reproduction, and behavior. Yet the neural and molecular systems that enable diverse organisms to adapt to these challenges remain understudied. Across vertebrates, the hypothalamus is central to organismal homeostasis and reproduction, while the hippocampus is critical for spatial behavior. To gain insight into the cellular and molecular plasticity mechanisms that afford neural systems resilience as they respond to ever-increasing environmental perturbations in a rapidly changing world, we examine two charismatic freshwater fish species – the colonially breeding Plains Longear Sunfish (Lepomis aequilensis) and the biparental Rio Grande Cichlid (Herichthys cyanoguttatus) – common across the longitudinal precipitation and latitudinal temperature gradients that

characterize the diverse eco-regions of Texas and surrounding regions. We use remote sensing and hydrological data to quantify environmental variability across >30 sites and find remarkable short- and long-term variability within and across sites. We then employ drone-based automated video tracking to quantify space use and reconstruct the social networks of these species, discovering that spatial and social behavior is remarkably resilient in even extreme environments. Finally, we use single nucleus RNA-sequencing of the fore- and midbrain to reveal the neuromolecular mechanisms underlying organismal resilience across highly variable environments. Taken together, our research integrates neurogenomics, behavior, and ecology to provide novel insights into the brain's ability to survive and thrive in highly variable and changing environments.

NEURO Fish integrative neurobiology in changing environments

Mapping Zebrafish Neural Activity Following Exposure to Increased CO2 and Temperature

Cordle, Jonathan (*jsc309@miami.edu*) Oral Presentation
Co-Authors: Heuer, Rachael and Ottervall, David
University of Miami

Abstract: Exposure to elevated CO_2 and temperature causes widespread impacts on behavior across freshwater and marine fish species, but the neural mechanisms underlying these observations remain incompletely characterized. Many studies point to alterations in neurotransmitter function, however neural responses likely reflect coordinated changes across multiple processes, particularly when stressors occur in combination. Assessing how neural activity varies across brain regions offers a broader framework for evaluating these responses and provides an opportunity to identify novel, region-specific alterations in neural function. Larval zebrafish are powerful vertebrate models for this approach, given the ease of imaging neural activity through their transparent head which can be paired with powerful customized genetic tools in subsequent studies. Distribution of neural activation can be mapped using established immunohistochemistry techniques known as MAP-Mapping to compare total Extracellular signal-Regulated Kinase (tERK) and its phosphorylated form (pERK), followed by confocal imaging, to generate 3D maps of neural activity in 294 distinct brain regions, many with known functions linked to specific behaviors. In this study, larval zebrafish were exposed to high CO_2, high temperature, and in combination, then behaviorally analyzed for visual startle response (VSR), acoustic startle response (ASR), and habituation for each. Following these assays, zebrafish were immediately preserved for MAP-Mapping analysis to establish links between environmental exposures, behavioral disruptions, and deviations in neural activation maps. Comparison of spatial activation between treatments may identify new mechanisms broadly applicable across species and help determine whether altered behavior in shifting environmental conditions is induced by localized dysfunction, or broader, brain-wide impacts.

NEURO Fish integrative neurobiology in changing environments

Characterization of environmental nitrite transport into the fish olfactory-brain axis

Restrepo, Nicole[1] (*nar116@txstate.edu*) Oral Presentation
Co-Authors: Casoli, Giulia[2]; Moretta, Mar[1]; Dharmasiri, Nihal[1], Bossi, Elena[2] and Huertas, Mar[1]
[1]Texas State University, [2]University of Insubria

Abstract: Anthropogenic activities release significant amounts of nitrogenous compounds into aquatic environments, including nitrite. Elevated levels of nitrite can be toxic, and its toxicity is linked to its accumulation in tissues.However, the mechanism by which nitrite is transported inside the tissues in aquatic vertebrates has not been characterized.We hypothesize that various exchangers/transporters from the solute carrier family are responsible for transporting environmental nitrite through the nose and gills of fish, where it accumulates in the brain and blood.
To test this hypothesis, we exposed goldfish to nitrite concentrations of 0 mM, 0.01 mM, 0.1 mM, and 1 mM for 26 days. At the end of the experiment, we collected gill and nasal samples to assess gene expression of several exchangers/transporters from the SLC26 and SLC15 families. Afterwards, nitrite transport was assessed by heterologous expression in Xenopus laevis of thecandidate transporters.
Gene expression analysis showed that nitrite induced a significant increase in slc15a4 in the nose, but significantly decreased expression in the gill. For slc26a3.2 and slc26a6l, there were no significant differences in concentration across treatments in either tissue type. Heterologous expression of slc15a4 in Xenopus oocytes showed transport of nitrite and nitrate. Analysis of SLC26 transporters is underway. In conclusion, the results of this study could be used to understand nitrogenous compounds transport and accumulation inside aquatic organisms. Furthermore, understanding nitrite transport could open new avenues of research into the use of nitrites at sublethal concentrations for therapeutic purposes.

NEURO Fish integrative neurobiology in changing environments

Exposure to three levels of CO2-induced freshwater acidification results in multi-tissue transcriptional and behavioural responses in medaka

Hasler, Caleb (*c.hasler@uwinnipeg.ca*) Oral Presentation
Co-Authors: Hutton, Melody; Minns, Rose and Jeffrey, Jen
The University of Winnipeg

Abstract: Elevated atmospheric carbon dioxide (CO2) can acidify aquatic environments, which several studies have linked to molecular and behavioural responses in fish. However, few studies on CO2-induced acidification have focused on freshwater fishes and the potential for fish to overcome acute responses. Our study investigated the effects of acidification over a 20 d exposure on medaka (Oryzias latipes) behaviour and mRNA transcripts sampled from the brain and gill. Adult medaka were exposed to three levels of acidification (6.1, 5.9, and 5.8 pH), observing behaviours on days 0, 5, 10, 15, and 20.

Brains were sampled on days 0, 5, 10, and 20 of pH treatments 6.1 and 5.8 to monitor relative abundance of mRNA of genes linked to GABA synthesis, reward-seeking behaviour, neurogenesis, feeding behaviour, circadian rhythm, and water and ion transport. Gill tissue was sampled in a similar fashion to explore mRNA abundance of genes related to ion transporters and acid-base regulation. Exposure to pH 5.8 altered some individual and group behaviours, including increased vertical activity and changes in zone preference on day-5. Sustained gill transcript responses occurred by day-5 of exposure, showing decreased relative mRNA abundance at 6.1 pH but increased abundance at 5.8 pH. Brain transcripts showed more delayed and selective adjustments. Our findings suggest that CO2-induced freshwater acidification causes some immediate behavioural responses in medaka, possibly linked to distinct, tissue-specific transcriptional pathways.

NEURO Fish integrative neurobiology in changing environments

Effects of environmental changes on the fish sense of smell: infections, pollution, and other horror stories.

Huertas, Mar (*mhuertas@txstate.edu*) Oral Presentation
Co-Authors: Billah, Maruf
Texas State University

Abstract: Smell has been one of the least appreciated senses throughout human history, frequently dismissed as communicating subjective feelings and linked to primary instincts. However, revolutionary advances in neuroscience since the mid-twentieth century have identified the sense of smell as a valuable model for studying the senses. The sense of smell is involved in the control of several physiological processes in vertebrates (e.g., reproduction, feeding, and behavior). Consequently, partial damage or complete loss of the sense of smell can negatively impact the health and survival of aquatic vertebrates. This presentation will summarize our research focused on two main sources of disturbances in the sense of smell, pollutants and pathogens, using fish as a model species. In the area of environmental pollutants, this talk will focus on the effects of aquatic nitrite on olfactory function. Nitrogen is a dangerous agricultural pollutant because it undergoes nitrification as a part of the nitrogen cycle, producing toxic nitrogenous compounds, such as ammonia, nitrate, and nitrite. We collected evidence showing that aquatic nitrite is neurotoxic and alters olfactory function in fish, with important implications for fish culture and fish well-being. In the context of pathogen detection, we showed that fish can detect pathogens and that the resulting olfactory signal is integrated across several brain areas via neurosteroid signaling to elicit an immune response. Our research aims to identify how environmental changes affect the senses and their long-term impact on vertebrate neurobiology.

NEURO Fish integrative neurobiology in changing environments

Changing environments affects neural nitric oxide pathways in fish

Moretta Urdiales, Maria Del Mar (*mariadelmarmorettaurdiales@gmail.com*) Oral Presentation
Co-Authors: Restrepo, Nicole and Huertas, Mar
Texas State University

Abstract: Nitric oxide (NO) is a neuromodulator in vertebrate sensory systems, including olfactory signaling. NO is produced via the aerobic pathway from L-arginine through the enzyme nitric oxide synthase (NOS), and via the anaerobic pathway from nitrite using reduced enzymes such as deoxyhemoglobin and xanthine oxidoreductase.Previous observations in our lab showed that fish exposed to non-lethal concentrations of nitrite have increased food attraction, suggesting that environmental nitrite may influence neural pathways involved in behavior. Our aim is to show that environmental nitrite accumulates in tissues and serves as a biological reservoir for NO synthesis, particularly under low-oxygen conditions where canonical NOS activity may be limited. For that reason, we exposed goldfish to several nitrite concentrations for 26 days and analyzed afterwards gene expression of neural NOS (nNOS), neuroglobin, and Xanthine reductase in the nose and gill. Moreover, we analyzed the in vivo and in vitro production of Nitrosylhemoglobin (HbNO) after exposure to various concentrations of nitrite and nitrate. Finally, we measured NO production in vitro in the nose and brain of goldfish exposed to nitrite. We observed reduced nNOS gene expression and decreased HbNO levels at increasing nitrite concentrations. The simultaneous decrease in HbNO and nNOS expression indicates that nitrite exposure triggers anaerobic NO production. Our investigation provides mechanistic evidence that environmental nitrite, which is increasing in our aquatic media due to agriculture run off, can act as a signaling precursorat sublethal concentrations, establishing a biochemical basis for nitrite-mediated modulation of neural and sensory function in fish.

NEURO Fish integrative neurobiology in changing environments

Thermal modulation of sensory-motor integration: developmental hyperthermia and chemical alarm cues in larval zebrafish (Danio rerio)

Vámos, Sofia (*sofia.duartevamos@ucdconnect.ie*) Oral Presentation
Co-Authors: Wollenberg Valero, Katharina
University College Dublin

Abstract: Temperature is one of the most prevalent environmental drivers shaping the physiology, development, and behavior of ectotherms. Early life stages are particularly sensitive to thermal conditions because organogenesis, neurodevelopment, and physiological systems are still forming, making embryos and larvae especially vulnerable to environmental stressors. We investigated how developmental hyperthermia (32°C) influences the sensory-motor integration of chemical stress cues (Stress Metabolites; SM) in larval zebrafish (Danio rerio). Using a high-throughput startle assay, we triggered the

Mauthner-cell-mediated escape circuit with acoustic-visual pulses. While larvae reared in 27°C control conditions exhibited significant sensory-motor plasticity with reduced startle magnitude when exposed to stress metabolites (SM, $p<0.001$), this behavioral sensitivity was absent in the 32°C larvae treatments. Larvae reared in thermal stress (TS) showed no significant difference in startle response between exposure to normal E3 media (TS) and stress-conditioned media (TSSM) ($p=0.74$), suggesting that developmental hyperthermia induces behavioral suppression to these cues. These data suggest that 32°C represents a functional threshold beyond which the Mauthner-mediated circuit loses/decreases its capacity for sensory integration. This decoupling indicates that chronic thermal stress limits behavioral plasticity, potentially driven by the disruption of excitatory signaling pathways. Our previous findings show that developmental hyperthermia changes the expression of NMDA receptor subunits (Grin1a/b) during early ontogeny, suggesting that thermal stress may impair the glutamatergic modulation required to calibrate M-cell excitability in response to environmental cues. Ultimately, a change in sensitivity to perceiving chemical cues implies altered responses of fish larvae to a changing environment.

NEURO Fish integrative neurobiology in changing environments

Rising to a challenge: Identifying the mechanisms mediating 'surfacing' behaviour in zebrafish

Gilmour, Kathleen (*kgilmour@uottawa.ca*) Oral Presentation
Co-Author: Tea, Michael
University of Ottawa

Abstract: Fish are considered to exhibit anxiety-like behaviour when they hug the sides of the tank in an open-field test, remain at the bottom of the tank in a novel tank diving test (NTDT), or prefer dark over well-lit areas of a tank. In zebrafish, exposure to selective serotonin reuptake inhibitors (SSRIs) has anxiolytic effects, with fish increasing time spent in the centre, top or well-lit area of a test tank. Zebrafish possess two paralogues of the serotonin transporter (Sert, slc6a4), Serta and Sertb. We exposed mutant zebrafish lines lacking functional expression of one or both Sert paralogues (serta-KOs, sertb-KOs, and sertab-KOs) to a battery of behaviour tests to evaluate the contribution of each paralogue to anxiety-related behaviour. Only sertab-KO fish exhibited anxiolytic behaviour in all tests. However, sertb-KO fish engaged in 'surfacing' behaviour in the NTDT, remaining at the top of the water column throughout the test, and this behaviour was also apparent in their housing tanks. We therefore hypothesized that surfacing behaviour was caused by serotonin-mediated hyperinflation of the swim bladder. Treatment of sertb-KO larvae with a tryptophan hydroxylase inhibitor to lower serotonin levels alleviated surfacing behaviour, and it was induced by treatment of wildtype fish with the SSRI fluoxetine to increase synaptic serotonin levels. As sertb expression is largely restricted to the hypothalamus of the brain, these data suggest a role for the hypothalamus in mediating swim bladder filling in zebrafish.

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The magic of mushrooms: Psilocybin influences behaviour in the mangrove rivulus, Kryptolebias marmoratus

Forsyth, Dayna (*dayna.f26@hotmail.com*) Oral Presentation
Acadia University

Abstract: Non-human animal models, including fish, are increasingly important for investigating how pharmacological agents such as hallucinogens influence behaviour, physiology, and cellular processes. These models help to reveal underlying mechanisms and support assessments of toxicological impact, efficacy, and safety. In this study, we used isogenic lineages of the amphibious mangrove rivulus (Kryptolebias marmoratus), an emerging model fish known for high activity and socially dynamic interactions. This species often display aggression towards conspecifics making it well suited to study behavioural responses to low doses of the psychoactive compound, psilocybin. We determined whether psilocybin could induce calming effects and reduce social aggression and activity. We socially stimulated fish using pairs of size-matched fish from different isogenic lineages and compared baseline social behaviour following a waterborne dose of psilocybin. Waterborne psilocybin treatment resulted in a significant decrease in activity levels and in the frequency of swimming bursts (an aggressive behaviour) towards a conspecific fish from a different lineage, with modest alterations on other behaviours. Our results also revealed considerable intraspecific variation in the behavioural response of these homozygous fish, suggesting the effects of psilocybin were largely independent of genotype. This study demonstrates that psilocybin reduces aggression and activity in an emerging fish model, adding to the evidence supporting psilocybin's potential as a therapeutic agent for future clinical translation.

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Trait Stability in the Open Field Test Predicts Selectively Bred Anxiety-like Phenotypes in Zebrafish

Johnson, Andréa[1] (*aljohns3@ualberta.ca*) Oral Presentation
Co-Authors: Hamilton, Trevor[2]; Mathot, Kimberley[1] and Hurd, Pete[1]
[1]University of Alberta; [2]MacEwan University

Abstract: Zebrafish (Danio rerio) are widely used in behavioural neuroscience to model anxiety-like behaviour due to conserved neural and stress axis mechanisms, yet substantial individual variability presents challenges for interpretation and reproducibility. Our previous longitudinal work using the novel tank dive test demonstrated stable individual differences in anxiety-related behaviour and identified high, medium, and low anxiety phenotypes based on zone preference. These findings support the presence of trait-like behavioural variation that likely reflects stable underlying neurobiological differences. The present study extends this framework using the open field test to characterise anxiety-like behaviour across development and to establish selectively bred anxiety lines. Adult zebrafish were assessed using time spent in the thigmotaxis zone alongside locomotor

measures, including swimming velocity and immobility. Individuals were classified as high anxiety (HAZ), medium anxiety (MAZ), or low anxiety (LAZ) based on thigmotaxis behaviour and used as founders for selective breeding. Behavioural and morphological outcomes were subsequently examined across generations. Results indicate that anxiety-related phenotypes identified in adulthood are stable, repeatable, and heritable. Larval assays revealed line-dependent differences in locomotor and exploratory behaviour, as well as morphological variation, suggesting early developmental divergence. Adult HAZ, MAZ, and LAZ fish exhibited distinct and consistent behavioural profiles in the open field test across generations. These findings demonstrate that anxiety-like behaviour in zebrafish can be reliably quantified, selectively propagated, and detected across life stages, providing a neurobiologically tractable model for investigating the mechanisms underlying stable individual variation in stress-related behaviour in fish.

NEURO Fish integrative neurobiology in changing environments

Climate-Driven Selection on Neurobiological Traits: Insights from Population Genomics

Rennison, Diana (*drennison@ucsd.edu*) Oral Presentation
Co-Authors: Wise, Caitlin and Ferranti, David
University of California San Diego

Abstract: Elucidating the genetic architecture of adaptation to climatic gradients requires understanding how environmental variation shapes biologically integrated traits, including those underlying neurobiological functions (i.e. behavior and sensory capacity). We surveyed hundreds of natural populations of the threespine stickleback (Gasterosteus aculeatus) distributed across multiple replicated climatic gradients spanning substantial variation in temperature regimes and seasonality. These gradients provide a powerful natural framework to test whether climate-mediated selection targets loci associated with neural function and neurobiological performance. We generated whole-genome resequencing data across populations and implemented covariance-aware genome–environment association analyses using BAYPASS to identify loci whose allele frequencies are significantly associated with climatic variables while accounting for shared demographic history. Across gradients, we detect a mixed genomic architecture characterized by both discrete regions of elevated differentiation and a widespread polygenic signal. Strikingly, climate-associated loci include genes involved in key neurobiological functions including genes central to sensory processing, suggesting that thermal and seasonal environments may shape adaptive variation in neural performance, behavior, and physiological integration. Patterns of parallelism across independent gradients further indicate that neurobiological pathways may represent repeatable targets of climate-driven selection. Together, these results highlight neuronal function as a key axis of climatic adaptation and reveal how polygenic changes contribute to evolutionary responses across heterogeneous environments.

NEURO Fish integrative neurobiology in changing environments

Neurogenetic mechanisms of behavioral resilience and susceptibility in larval fish within changing environments

Tatarsky, Rose (*rtatarsky@ucsd.edu*) Oral Presentation
Co-Authors: Lovett-Barron, Matthew; Rennison, Diana and Tresguerres, Martin
University of California San Diego

Abstract: Animals rear offspring in dynamic landscapes where larvae must navigate complex sensorimotor challenges. This transition is critical for anadromous three-spined stickleback; larvae move from protected nests and paternal care to fluctuating habitats as they develop toward their migration from freshwater nurseries to marine environments. By leveraging divergent stickleback populations adapted to distinct ecological niches, we can investigate conserved neurogenetic mechanisms of resilience supporting larval survival during this "nest-to-dispersal" window. This provides a powerful model for predicting how larval behaviors may adapt or fail in rapidly changing environments.
Using high-throughput tracking (SLEAP), we established a quantitative behavioral timeline from hatching through the juvenile stage, discovering that marine stickleback larvae coordinate their swimming to school shortly after hatching. We are probing the sensory basis of this uncharacterized larval behavior via light/dark trials to distinguish the relative contributions of visual and mechanosensory integration.
To map the underlying functional neuroanatomy, we developed Hybridization Chain Reaction (HCR) probes to visualize cfos and neuromodulatory activity within putative sensory integration centers. We will compare behavior and baseline neural maps against larvae under acute thermal stress, hypoxia, and acidification to pinpoint windows of neurodevelopmental resilience or susceptibility to these environmental changes. In parallel, we are developing CRISPR/Tol2-mediated GCaMP transgenesis for real-time neuroimaging while utilizing Genome-Wide Association Mapping (GWAS) to link candidate genes with these resilient and susceptible phenotypes. By linking behavior to neural and genetic markers, we aim to better predict how larval fish broadly will respond to the physiological demands of a rapidly changing climate.

NEURO Fish integrative neurobiology in changing environments

Neurobiological responses to thermal pressure in natural threespine stickleback populations

White, Matthew (*m8white@ucsd.edu*) Oral Presentation
Co-Authors: Wirth, Taylor; Martz, Todd; Rennison, Diana; Lovett-Baron, Matthew and Tresguerres, Martin
University of California

Abstract: Nervous systems allow animals to interact with their environments, ensuring appropriate sensory responses in dynamic diel, seasonal, and anthropogenic conditions. What neurobiological activity confers adaptation to changing environments and how could this provide resilience to anthropogenic pressure? I am adapting neuroscience tools,

including whole-brain imaging and quantitative behavior paradigms, from the zebrafish model system to study natural populations of threespine stickleback fish in an ecologically relevant context. I am currently using these tools to explore the neural activity and gene expression changes underlying behavioral responses to an altered thermal environment. I will discuss recent work to record free-moving and head-tethered behavior at distinct temperatures and report the corresponding neural activity patterns. I will also discuss future efforts to probe sensory systems and establish genetic tools in stickleback.

NEURO Fish integrative neurobiology in changing environments

Structure-function relationships of the rainbow trout visual system following environmental hypoxia

McKinley, Abigayle (*mckinlea@uoguelph.ca*) Oral Presentation
Co-Author: Turko, Andy and Bernier, Nick
University of Guelph

Abstract: Vision is an important sensory modality in fishes, but the retina also has a high energetic cost, making it potentially vulnerable to limitations in oxygen availability. Although human activities are increasing the frequency and severity of hypoxic events in aquatic ecosystems, the effects of hypoxia on the visual system are not well understood. Retinal neurons in rainbow trout (Oncorhynchus mykiss) are vulnerable to hypoxia in vitro, but in vivo, specialized anatomy such as the choroid rete mirabile may maintain retinal oxygen tension during environmental hypoxia. Our study uses an integrative approach to evaluate the relative vulnerability and resilience of the visual system by quantifying structural, molecular, and functional responses following hypoxia exposure. We found that acute hypoxia induced a delayed thinning of the retinal pigment epithelium, an energetically expensive retinal support structure, while the neural layers remained structurally intact. Furthermore, fish exposed to chronic hypoxia demonstrated impaired visual tracking at an intermediate spatial frequency using an optomotor response assay, despite maintaining responses at low-frequency stimuli. To further investigate the detrimental effects of hypoxia and the potential for regeneration, we are characterizing the expression of genes associated with apoptosis and cellular proliferation. By linking histological and molecular properties to behavioral outcomes, this research provides novel insights into the mechanisms that contribute to retinal plasticity and the vulnerability and resilience of the visual system of fishes to the rising incidence of environmental hypoxia.

NEURO Fish integrative neurobiology in changing environments

Teleost cerebrospinal fluid response to future ocean conditions

Kwan, Garfield[1] (*gkwan09@gmail.com*) Oral Presentation
Co-Author: Finlay, Jennifer[1]; Johnson, Andréa[2]; Donnelly, Aidan[1]; Jarvis, Jack[1]; Sanders, Trystan[1]; Savin, Oleg[3]; Hamilton, Trevor[3]; Tresguerres, Martin[4] and Wilson, Rod[1]
[1]University of Exeter, [2]University of Alberta, [3]MacEwan University, [4]University of California San Diego

Abstract: Future ocean conditions include changes in temperature, CO2/pH, O2, and food availability – all of which can alter cerebrospinal fluid (CSF) ionic and acid-base chemistry, and potentially modify fish behavior. For instance, Nilsson et al. (2012) hypothesized fish behavioral impairment associated with ocean acidification is linked to physiological adjustments of CSF chemistry, specifically increased [HCO3-] and decreased [Cl-] that can reverse GABAA receptor's neuronal electrochemical potential (EGABA). This talk focuses on the CSF and intracellular brain chemistry in teleost fishes, and will include Atlantic lumpfish (C. lumpus) response to 1-hour, 5-day, and 5-weeks of high CO2 (3,000 – 6,000 µatm), and European seabass (D. labrax) response to 2, 6, and 12-weeks of multi-stressor exposure: high CO2 (3,500 µatm) x hypoxia (65% DO), or temperature (18 – 26°C) x rationing (3% body weight once or thrice per week). Lumpfish CSF pH was maintained and CSF [HCO3-] accumulated – but CSF [Cl-] was not affected by CO2 exposure. Combined with respective intracellular brain pH, [HCO3-], and [Cl-], we did not detect EGABA reversal in the lumpfish. European seabass CSF pH and [HCO3-] responded similarly to high CO2 independent of hypoxia, whereas cooler temperature and fasting increased CSF pH. Analysis of European seabass ionic and brain intracellular chemistry is ongoing, and together with CSF data will determine whether future ocean conditions could reverse EGABA. This study highlights the need for direct CSF measurements due to species- and fluid-specific responses, and it advances a new toolkit to investigate CSF regulation and neurobiology in non-model fishes.

NEURO Fish integrative neurobiology in changing environments

Hidden in the Vehicle: Behavioural Effects of Solvent Exposure in Larval Zebrafish

Hagen, Ethan[1] (*evhagen@ualberta.ca*) Poster Presentation
Co-Authors: Harper, Matthew[2]; Zhang, Yanbo[1]; Hamilton, Trevor[2]
[1] University of Alberta; [2] MacEwan University

Abstract: Zebrafish (Danio rerio) are frequently used to study the behavioural impact of toxicological compounds. Larval zebrafish continue to gain interest as a model due to their high-throughput nature and extensive behavioural measures, allowing for rapid and effective testing. Often, solvents like dimethyl sulfoxide (DMSO), methanol, or ethanol are used, but their sub-lethal impact on larval zebrafish behaviour is not yet well defined. Here, we investigate the impact of these three solvents at 0.01%, 0.1%, and 1.0% vol/vol on locomotion and startle responses following a 30-minute exposure in 5-day-old larval zebrafish. We found no effect of DMSO on distance moved in a spontaneous swim test

compared to controls and no effect on dark startle, light startle, or tap startle responses. Both methanol and ethanol at 1.0% increased the total distance moved compared to controls, and ethanol at 1.0% decreased the dark startle response. Methanol and ethanol did not alter the light startle or tap startle response at the tested doses. The results of this study suggest that DMSO is the best solvent to use to minimize confounding behavioural alterations, followed by methanol, and finally ethanol.

NEURO Fish integrative neurobiology in changing environments

Prolonged exposure to perfluorooctanesulfonic acid (PFOS) exposure alters anxiety-like behaviour but not boldness in zebrafish (Danio rerio)

Kutcher, Ashley[1] (*kutchera@mymacewan.ca*) Poster Presentation
Co-Authors: Hamilton, Trevor[1]; Hurst Radke, Nicole[2]; Szaszkiewicz, Joshua[1]; Nadolski, Nathan[2]; Franczak, Brian[1]; Ross, Matthew[1]; Velupillai, Niru[1] and Legge, Eric[1]
[1]MacEwan University, [2]University of Alberta

Abstract: Perfluorooctanesulfonic acid (PFOS) is an environmental contaminant with toxic properties that are resistant to breakdown, leading to bioaccumulation in aquatic life, humans, and the environment. PFOS can produce toxic effects in zebrafish, such as altered immune system-related intestinal functions, altered gene expression and increased energy expenditure. However, the impact on relatively complex behaviours such as boldness and anxiety-like behaviour is unknown. To investigate PFOS's effect on behaviour, we exposed adult zebrafish to either 0, 0.1, or 1.0 mg/L of PFOS for 40 days, and used motion-tracking software to assess locomotion, anxiety-like behaviour, and boldness.Testing was conducted after 20- and 40-days with individual testing in an open field arena, novel approach test and novel tank dive test.We observed a significant decrease in anxiety-like behaviour with 40-days of PFOS exposure at 1.0 mg/L, with no differences in locomotion or approach to the novel object.These results suggest that chronic PFOS exposure has sub-lethal impacts on behaviour that warrant further investigation.

NEURO Fish integrative neurobiology in changing environments

Disruption of endocannabinoid signalling during gastrulation with 9 tetrahydrocannabinol (THC) or cannabidiol (CBD) alters γ aminobutyric acid (GABA) expression in developing zebrafish (Danio rerio)

Mamo, Grace (*gmamo@ualberta.ca*) Poster Presentation
Co-Author: Ali, Declan
University of Alberta

Abstract: Early embryonic development in teleost fishes involves neural patterning and the establishment of inhibitory and excitatory balance, which is required for normal larval physiology, including sensory processing, motor control, and behaviour. Gastrulation is a vital developmental stage during early development in which body axes are specified, and

the basic organization of major organ systems are established. The endocannabinoid system (ECS) is present early in zebrafish development and functions as a conserved neuromodulatory signalling system involved in neural differentiation and synaptic organization. To examine the contribution of ECS signalling to zebrafish neurodevelopment (specifically during gastrulation), D. rerio embryos were exposed during gastrulation to a vehicle control or to exogenous cannabinoids (THC or CBD) for the duration of gastrulation. Once the larvae reached 5 dpf (days post fertilisation), immunohistochemistry was used to assess the distribution of γ-aminobutyric acid (GABA) and vesicular glutamate transporters 1/2 (VGLUT1/2). Preliminary data show that THC exposure during gastrulation produces a dose-dependent reduction in GABA immunoreactivity in the brain. Conversely, CBD exposure causes a dose-dependent increase in GABA immunoreactivity. The effects observed with these exposures are attenuated by co-exposure with cannabinoid receptor blockers, which indicates that ECS receptor-mediated signalling is likely responsible for the effects seen in the exposure groups. Changes in VGLUT1/2 expression in THC or CBD exposure groups are less pronounced and remain variable across treatment groups; these effects appear modest relative to those observed for GABA and should be considered preliminary.

NEURO Fish integrative neurobiology in changing environments

Single-nucleus RNA-sequencing of the hypothalamus reveals the molecular underpinnings of physiological and social homeostasis across variable environments

Phofolos, Demetrius (*dap4688@my.utexas.edu*) Poster Presentation
Co-Author: Hofmann, Hans
The University of Texas

Abstract: Highly variable environments expose animals to large, and often sudden, fluctuations in temperature, precipitation, and other climatic factors, which can pose a potent threat to the maintenance of physiological and social homeostasis. Still, countless species survive, and even thrive, in highly variable environments. For example, males of the Plains Longear Sunfish (Lepomis aequilensis) form fiercely defended nesting colonies across the dramatic latitudinal temperature and longitudinal precipitation gradients of Texas. The vertebrate hypothalamus is central to organismal homeostasis and social behavioral regulation, yet the molecular and cellular mechanisms that govern how animals achieve resilience within their natural habitats are woefully understudied. Additionally, our characterization of homeostatic neuronal populations is limited by a poor understanding of their underlying gene co-expression architecture. To better understand and predict the strategies animals use to withstand environmental variability, we combined climatological and hydrological data with single-nucleus RNA-sequencing of the hypothalamus of reproductive L. aequilensis males across sites in Central Texas. We first examined the composition of homeostatic neuronal populations at the level of transcriptional programs. Next, we investigated how the transcriptomes of these neurons conferred resilience within and across highly variable environments, in real time and over the course of seasons and decades. Our results begin to uncover the homeostatic transcriptional programs that

regulate reproduction, energy homeostasis, timekeeping, stress response, temperature-sensing, and osmoregulation in the face of environmental variance across space and time.

NEURO Fish integrative neurobiology in changing environments

The anxiety-reducing drug buspirone has differential effects on selectively bred zebrafish at larval and adult stages

Steman, Tobin[1] (*t.steman@icloud.com*) Poster Presentation
Co-Authors: Johnson, Andréa[2] and Hamilton, Trevor[1]
[1]MacEwan University; [2] University of Alberta

Abstract: Zebrafish (Danio rerio) are a popular model organism for pharmacological compound research and for assessing the impact of wastewater runoff. Here, behavioural effects of the 5-HT1A agonist buspirone were examined in selectively bred low- (LAZ), moderate- (MAZ), and high-anxiety (HAZ) zebrafish in both larval and adult stages. Buspirone exposure was assessed in larvae at 5 days post-fertilization (150 µM, 30 minutes) through open field spontaneous swimming and dark (DSS), light (LSS), and tap startle stimuli (TSS) response tests. Adults were tested at 7 months old (10 and 100 mg/L, 10 minutes) using open-field (OF) and novel object approach (NOA) tests. Buspirone did not significantly affect larval distance moved during spontaneous swimming, suggesting no sedative effect. Immobility (freezing) was affected during both LSS and TSS, with buspirone reducing MAZ and HAZ freezing while increasing LAZ freezing. In the adult OF and NOA tests, distance moved and time spent highly mobile were the most reduced in HAZ fish at the low (10 mg/L) buspirone dose. In contrast, the high (100 mg/L) dose increased immobility and reduced locomotion in all groups, with HAZ most strongly affected. Adult MAZ treated with the high dose remained in the centre zone with the novel object longer than with the low dose and compared to LAZ and HAZ fish administered the high dose. This study suggests that zebrafish with different behavioural profiles respond to pharmacological compounds with varying outcomes, with implications for responsivity to wastewater effluent containing pharmaceuticals.

CHANGE I Integrating physiology and behaviour in a changing environment

Integrating physiology and behaviour in a changing environment

Bradley Howell, Caleb Hasler, Nann Fangue and Steve Cooke

Abstract: This symposium will explore the growing body of research connecting physiological processes with behavioural outcomes in fishes across ecological contexts. From metabolism and energetics to stress and neuroendocrine function, internal states shape how fishes forage, migrate, interact socially, and respond to environmental change. By bringing together researchers studying diverse species and systems in both field and lab settings, this symposium will highlight how integrating physiology, and behaviour can illuminate adaptive strategies, inform conservation, and advance predictive models of animal performance. Building on past ICBF symposia that emphasized integrative biology and environmental physiology, this session aims to bridge mechanistic insights with ecological relevance. We will spotlight advances in methodologies—such as biologging, acoustic telemetry, behavioural assays, respirometry, and "-omics"—that enable researchers to track fine-scale behavioural variation in relation to internal physiological states. In doing so, the symposium will examine how these individual-level insights can reveal broader patterns in phenotypic plasticity, transgenerational acclimation, population dynamics, species interactions, and ecosystem responses to global change.

CHANGE I Integrating physiology and behaviour in a changing environment

Mechanistic conservation of wild fishes demands integration of animal physiology and behaviour

Cooke, Steven J. (*steven_cooke@carleton.ca*) Oral Presentation
Carleton University

Abstract: In this presentation I argue that the integration of behaviour and physiology when engaging in mechanistic conservation research of wild fishes isn't just a "nice to have" but is essential. In the wild it is nearly impossible to decouple behaviour and physiology. Animal-environment interactions and animal-human interactions are a manifestation of behaviour and physiology - with complex interconnections and feedbacks. I will use a range of examples to demonstrate why and how such integration makes sense.

CHANGE I Integrating physiology and behaviour in a changing environment

Hot and bothered: behavioural and physiological responses to warming in freshwater fish

De Bonville, Jeremy (*jeremy.debonville@gmail.com*) Oral Presentation
Co-Authors: Raby, Graham
Trent University

Abstract: In freshwater ecosystems, increasingly frequent heatwaves threaten the physiological performance, behaviour, and survival of ectotherms such as fish. The ability of fish to acclimate to fluctuating temperatures and the metrics used to quantify it are central to predicting resilience and guiding conservation efforts. The critical thermal maximum (CTmax), the temperature at loss of equilibrium during warming, is the most common measure of upper thermal tolerance as fish typically survive the trials. However, CTmax may lack ecological realism since wild fish rarely encounter such rapid warming and often have the opportunity to escape before temperatures become lethal. A complementary behavioural metric, the agitation temperature (Tag), occurs several degrees below CTmax and represents the temperature at which fish begin to avoid heat or display escape behaviour. Conceptually, Tag bridges physiological and behavioural thresholds, yet its repeatability, plasticity and links to physiological metrics remain unexplored. To assess the plasticity of both traits, we tested how Tag and CTmax responded to thermal acclimation in two freshwater fish species: bluegill (Lepomis macrochirus) and largemouth bass (Micropterus nigricans). Bluegill exhibited slightly higher tolerance to warming for both traits. We then evaluated repeatability of Tag and CTmax in bluegill across acclimation temperatures by testing individuals three times. While CTmax was repeatable, Tag showed greater inter-individual variability, suggesting that behavioural thermal thresholds may be more flexible. Understanding the repeatability and plasticity of Tag will help determine its validity as a behavioural analogue to CTmax and its potential use in predicting fish responses to thermal stress.

CHANGE I Integrating physiology and behaviour in a changing environment

Does temperature variability change how we interpret climate resilience in a nocturnal reef shark?

Emmons, Sophia M.[1] (*sophia.emmons@my.jcu.edu.au*) Oral Presentation
Co-Authors: Benoit, Aurélie[1]; Humphrey, Craig[2]; Simpfendorfer, Colin[1]; Seibel, Brad[3] and Rummer, Jodie[1]
[1]James Cook University, [2]Australian Institute of Marine Science, [3]University of South Florida

Abstract: Most studies investigating the impacts of climate change on aquatic animals rely on static temperature treatments that do not reflect the dynamic environments many species inhabit. On shallow reef flats, daily temperatures commonly fluctuate by >5°C., exposing resident fishes, like juvenile and adult epaulette sharks (Hemiscyllium ocellatum), to predictable diel cycles. Epaulette sharks exhibit clear circadian patterns in metabolism and behaviour, resting during the day and foraging at night. Understanding how physiological performance responds to natural thermal variability is therefore essential for interpreting ecological resilience. We simulated ecologically realistic seasonal diel temperature cycles based on reef flat conditions from Heron Island (southern Great Barrier Reef) using 24 epaulette sharks maintained at the National SeaSimulator (Australian Institute of Marine Science). Over eight weeks, sharks experienced diel temperature fluctuations representative of winter (n=8, 23-28°C) or summer (n=8, 28-33°C), or a static

summer temperature control (n=8, 28ºC). Metabolic rates were estimated across key activity and temperature periods using intermittent-flow respirometry, focusing on the shared temperature of 28ºC among treatments. We also quantified haematological indicators of energy use, including haemoglobin, haematocrit, mean corpuscular haemoglobin concentration (MCHC), lactate, glucose, and b-hydroxybuterate. Oxygen transport traits were similar between winter and control treatments, whereas, sharks in the fluctuating summer treatment exhibited significantly higher haematocrit and lower MCHC. Glucose increased stepwise with temperature, suggesting greater reliance on anaerobic metabolism with warming, regardless of diel thermal variation. Incorporating natural temperature variability provides a more ecologically relevant framework for interpreting physiological performance and climate resilience in reef sharks.

CHANGE I Integrating physiology and behaviour in a changing environment

Evaluating climate change resilience in wild white sturgeon populations

Earhart, Madison[1] (*earhart@zoology.ubc.ca*) Oral Presentation

Co-Author: Bouyoucos, Ian[1]; Bugg, William[1,2]; Piczak, Morgan[1]; Scott, Kelly[3]; Kussin-Bordo, Noah[4]; Schrier, Sarah[5]; Brauner, Colin J.[1]; Schulte, Patricia[1]; Eliason, Erika[6] and McAdam, Steve[7]

[1]University of British Columbia, [2]Pacific Salmon Foundation, [3]Tsawwassen First Nation, [4]Lower Fraser Fisheries Alliance, [5]Fraser River Sturgeon Conservation Society, [6]Department of Fisheries and Oceans Canada, [7]Ministry of Water Land Resources Stewardship

Abstract: The ability to understand and predict the impacts of human-altered environments on aquatic animals is critical in order to create impactful conservation strategies and management plans, particularly for species at risk. Sturgeon are prehistoric, long-lived fishes that make up one of the most endangered groups of fishes globally with species distributed across the Northern Hemisphere. White sturgeon (Acipenser transmontanus), the largest and most ancient freshwater fish in North America, are threatened throughout their range in British Columbia (BC) including populations in the Fraser River (threatened) and Nechako River (endangered), which face a variety of anthropogenic stressors. The population declines in these rivers are concerning to rights and title holders (First Nations) and shareholders throughout BC. In order to aid the recovery of white sturgeon we must identify the causes of population declines, low juvenile recruitment, and mortality events. To do this, we created novel mRNA biomarker panels that assess climate change stressor resilience, or lack thereof, in wild sturgeon populations. Specifically, we have assessed physiological status and the impacts of climate warming, food limitation, and pathogen burden on hundreds of wild sturgeon in multiple BC populations through minimally invasive sampling and community science. Results will be discussed in the context of yearly and seasonal effects of climate stressors and pathogen abundance on wild sturgeon resilience, metabolic rate, growth rate, habitat use, and recreational angling impacts. Our data will be used to inform ongoing white sturgeon recovery efforts prioritizing co-management of this iconic species.

CHANGE I Integrating physiology and behaviour in a changing environment

Does tolerance to fast warming predict tolerance to slow warming in fishes?

Lechner, Emily[1] (*emily.lechner@bioenv.gu.se*) Oral Presentation
Co-Author: Stewart, Erin[2]; Jutfelt, Fredrik[1]; Cowan, Zara[1]; Ekelin, Iggy[1]; Hoots, Elizabeth[3]; Kuchenmueller, Luis[3]; Blewett, Tamzin[4]; DeBonville, Jeremy[2]; Binning, Sandra[5]; Raby, Graham[2]; Roche, Dominique[6]; Gradito, Maryane[3]; Tamarit, Elena[1]; Vigneault, Juliane[7]; Melancon, Vincent[5]; Clark, Timothy[3]; Ern, Rasmus[8]; Bihun, Christian[2]; Cadonic, Ivan[4]; Metz, Moa[8]; Tomas, Mafalda[1]; Pfeufer, Leon[1] and Pottier, Patrice[1]
[1]University of Gothenburg; [2]Trent University, [3]Deakin University, [4]University of Alberta, [5]Université de Montréal, [6]Université de Neuchâtel, [7]Université du Québec à Rimouski, [8]Norwegian University of Science and Technology

Abstract: Critical thermal maximum (CTmax) is widely used to quantify upper thermal limits in fishes, but its ecological relevance is often questioned because it is typically measured using rapid heating rates (commonly ~0.3°C min^{-1}). In natural environments, fish more frequently experience thermal stress under slower warming over hours to days. For climate vulnerability assessments and mechanistic species distribution models, it is therefore important to determine whether CTmax values obtained under fast warming predict tolerance under slower, more ecologically realistic rates, or whether different physiological processes dominate.
Here, we quantifiedindividual-levelcorrelationsof upper thermal tolerance under fast (0.3°C min^{-1}) and slow (1°C h^{-1}) warming in seven temperate fish species. Weobservedsubstantial interspecific variation in both themagnitudeand direction of differences between warming rates.
Across species,CTmaxvalues measured under fast and slow warming wereweaklycorrelated,andsignificant correlations were detected in only three of seven species. Our resultsindicatethatCTmaxmeasured under standard rapid ramping protocols cannot be assumed to reliably predictindividual or speciestolerance to slower, ecologically realistic warming across fish species. This heterogeneitymayreflectdifferences in acclimation capacity and damage–repair dynamics during heat stress.
WhileCTmaxremainsa valuable tool, estimates obtained under different warming rates may need to be matched to specific ecological and modelling applications.

CHANGE I Integrating physiology and behaviour in a changing environment

Physiological and genomic evidence of differential thermal performance in Kokanee salmon (Oncorhynchus nerka)

Mayer, Noa[1] (*noabermanmayer@gmail.com*) Oral Presentation
Co-Authors: Eliason, Erika[2]; Hinch, Scott[1]; Miller, Kristi[2] and Russello, Michael[1]
[1] University of British Columbia, [2] Fisheries and Oceans Canada

Abstract: Pacific salmon (Oncorhynchus spp.) are particularly vulnerable to climate-induced warming due to narrow thermal optima, exposure to elevated temperatures during rearing and migration, and limited capacity to extend upper thermal tolerance. For the first

time, we quantified aerobic scope (AS), critical thermal maxima (CTmax), genomic stress signatures, and infectious agent presence and burden across a thermal gradient (12–24 °C) in three Kokanee salmon (O. nerka) populations (Coldstream, Mission, Norbury). Marked inter-population differences in thermal performance were observed. Coldstream fish experienced 100% mortality at 22 °C, Mission at 24 °C, whereas Norbury fish maintained greater tolerance (50% mortality at 24 °C). Maximum metabolic rate declined sharply at elevated temperatures in Coldstream, resulting in significantly reduced absolute AS relative to Norbury and Mission ($p < 0.001$). Coldstream also exhibited higher gill infectious agent burden and elevated transcriptional signatures of thermal and hypoxic stress, while Norbury maintained greater aerobic capacity and lower stress-associated gene activation across temperatures. Integration of immune gene expression profiles into metabolic models revealed a significant negative relationship between activation of an innate-dominated immune response and absolute AS ($p = 0.027$). Individuals exhibiting higher innate immune stimulation consistently displayed reduced absolute AS across temperatures, and inclusion of this variable significantly improved model explanatory power beyond temperature × population effects alone. Together, these results highlight how interacting physiological, genomic, and disease processes influence thermal tolerance, providing new insight into the mechanisms underlying population-specific vulnerability to climate-driven warming.

CHANGE I Integrating physiology and behaviour in a changing environment

Measuring thermal tolerance and metabolic recovery times of wild salmonids after high thermal exposures: Implications for angling policies in Atlantic Canadian river systems

Morash, Andrea (*amorash@mta.ca*) Oral Presentation
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Abstract: Thermal variability is common in most aquatic environments, and is expected to increase along with extreme thermal events as a result of climate change. However, the potential effects of this variability have not yet been fully incorporated into policies for fish conservation. For example, the current “warm water protocol” policy on angling of wild salmonids in Atlantic Canada closes the fishery when water temperatures remain above 20°C for 2 consecutive days, and re-opens the fishery after 2 consecutive nights of <20°C. However, this policy does not consider daily maximum temperatures in the preceding days, and there are currently no measurements of recovery status in these fish to ensure that this timeline is appropriate. To better understand thermal tolerance and recovery times, we acclimated wild-caught Brook char to a summer thermal profile of 16-21°C based temperatures commonly observed in the Miramichi River. We exposed these fish to and environmentally relevant increase in maximum temperature and measured whole-animal metabolic rate, thermal tolerance using an ecologically relevant critical thermal swimming test, recovery time, and a suite of metabolites immediately after the thermal exposure and after full recovery.We found that measuring thermal tolerance under more realistic

conditions yields a significantly lower thermal tolerance (24°C vs. 31°C) than traditional critical thermal maximum tests. In addition, we found that complete metabolic recovery can take up to 3-4 days even when held under ideal cool conditions (16°C). These data suggest that current policies on angling may not be sufficient to protect salmonids during warm thermal exposures.

CHANGE I Integrating physiology and behaviour in a changing environment

Habitat thermal variability predicts upper thermal tolerance in a coastal triplefin fish

Johnson, Sheri (*sheri.johnson@otago.ac.nz*) Oral Presentation
University of Otago

Abstract: The Climate Variability Hypothesis predicts that organisms from more thermally variable environments should exhibit greater tolerance to temperature extremes than those from more stable habitats. We tested this prediction in New Zealand triplefins of the genera Forsterygion and Bellapiscus, quantifying critical thermal maximum (CTmax) in fish collected from estuarine, harbour, rockpool, intertidal and subtidal habitats that differ in thermal exposure. Fish were subjected to a standardized warming protocol to determine upper thermal limits and were monitored following trials. CTmax differed among habitat groups, with individuals from more thermally variable environments exhibiting higher upper thermal limits than those from comparatively stable habitats, consistent with predictions of the Climate Variability Hypothesis. Although most fish recovered following assays, the common triplefin, a generalist that transitions between intertidal and subtidal environments, exhibited substantial delayed mortality within 24 hours of testing, whereas other groups showed little or no mortality. Ongoing analyses are examining whether behavioural responses during warming, including agitation temperature—the onset of heightened activity prior to loss of equilibrium—and changes in ventilation rate provide earlier indicators of thermal stress than CTmax alone. These findings suggest that fine-scale environmental differences can shape thermal tolerance and that acute thermal limits may not fully capture post-stress survival. Understanding such variation will improve predictions of how small coastal fishes respond to increasing thermal variability under climate change.

CHANGE I Integrating physiology and behaviour in a changing environment

Linking metabolism, physiological performance and oxygen transport in Baltic cod exposed to prolonged mild hypoxia

Husen, Magnus H.[1] (*mhhu@dtu.dk*) Oral Presentation
Co-Authors: Behrens, Jane W.[1]; Birnie-Gauvin, Kim[1]; Andreasson, Anna H.[1]; Brander, Keith[1]; van Deurs, Mikael Seehusen Kruchov[1]; Andersen, Niels Gerner[1]; Sørensen, Sune Riis[1] and Fago, Angela[2]
[1]Technical University of Denmark, [2]Århus University

Abstract: Oxygen depletion is increasing globally, particularly in semi-enclosed areas such as the Baltic Sea. Fish can behaviorally avoid hypoxic waters but the expanding spatial and temporal extent of hypoxic zones, especially during summer and autumn months, forces individuals to occupy suboptimal conditions. This is especially true for the Baltic cod (*Gadus morhua*), which delve into deeper waters characterized by reduced oxygen availability to avoid predators and forage on benthic prey. Despite its ecological relevance, the physiological consequences of long-term exposure to mild hypoxia remain poorly understood. Here, we experimentally investigated how prolonged exposure to mild hypoxia (~51% air saturation, corresponding to approx. ~5 mgO2 l-1) over six weeks affects metabolism (MO2), oxygen transport, digestive capacity and growth performance in Baltic cod fed natural prey ad libitum. We demonstrate that decreasing oxygen availability to what is considered a mild level of hypoxia has significant negative effects on several key fitness-related traits. This was despite the fish in mild hypoxia having increased hemoglobin levels and hemoglobin-oxygen affinity, facilitating improved oxygen binding at the gills and augmenting oxygen carrying capacity. Yet these physiological adjustments were clearly insufficient to mitigate the adverse effects of prolonged mild hypoxia on aerobic scope. Our findings demonstrate that persistent mild hypoxia can generate cascading impacts on individual performance and growth and likely has contributed to the observed long-term, drastic decline in Baltic cod size and nutritional condition.

CHANGE I Integrating physiology and behaviour in a changing environment

No air, no action: cod hit the slow lane under hypoxia

Birnie-Gauvin, Kim (*kbir@aqua.dtu.dk*) Oral Presentation
Co-Authors: Husen, Magnus; Behrens, Jane W.; Andreasson, Anna H. and van Deurs, Mikael Seehusen Kruchov
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Abstract: Atlantic cod (Gadus morhua) is under severe pressure across most of its range. In the Baltic Sea, cod size at maturity and spawning success have spiraled down to an all-time low despite decades of intensive management efforts. Aside from fishing pressure, one of the primary factors thought to impact cod is low oxygen levels. Many studies have showed how hypoxia can reduce metabolism and growth in fishes, but relatively few have explored how activity may be influenced by low oxygen levels. Here, we tagged cod with tri-axial accelerometers and exposed some of them to mild hypoxia (50% air saturation, 5

mgO2 l-1) for an extended period of time (seven weeks). Over this period, we collected information on tailbeat frequency (TBF) and vectorial dynamic body acceleration (VeDBA). We linked these activity metrics to overall activity levels, feeding events and a stress response test. Our findings unequivocally show that hypoxia-exposed cod hit the slow lane: activity levels were generally lower, with lower TBF and VeDBA values throughout the experiment. Activity levels following feeding and the escape response test were also lower in hypoxia-exposed fish than those under normoxia. Our findings contribute to our understanding of the effects of prolonged hypoxia on fishes and suggest that previously-shown effects of hypoxia on physiology translate to behavioural effects. It remains unknown whether these behavioural changes could affect foraging ability, predator escape or spawning success, though this would align with the findings of Husen et al., showing reduced growth in hypoxia-exposed cod.

CHANGE I Integrating physiology and behaviour in a changing environment

Developmental neurobehavioural toxicity of Bisphenol F in zebrafish larvae

Hasan, A. K. M. Munzurul (*munzurul.hasan@usask.ca*) Oral Presentation
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University of Saskatchewan

Abstract: Bisphenol F (BPF), a structural analogue of bisphenol A, increasingly used in industrial and consumer products, has been widely detected in aquatic environments. Despite being marketed as a safer alternative, its developmental neurotoxicity remains poorly understood. This study investigated whether embryonic exposure to environmentally relevant concentrations of BPF alters early neurodevelopment and behaviour in zebrafish (Danio rerio) and explored underlying molecular mechanisms. Embryos were exposed to 0, 5, 20, and 50 µg/L BPF from 4-20 hours post-fertilization (hpf). Spontaneous tail-coiling was assessed at 20 and 30 hpf, and at 120 hpf, larvae underwent thigmotaxis, dark/light preference, and locomotor activity tests. BDNF protein and neuroplasticity- and stress-related gene expression were analyzed in whole larvae. BPF did not alter spontaneous tail-coiling but induced significant behavioural changes at 120 hpf. Exposed larvae showed anxiety-like behaviour, evidenced by reduced time and distance in the outer zone during thigmotaxis. Reflexive movement increased at all concentrations, particularly 20 µg/L, indicating heightened neural excitability. In the dark/light test, locomotor activity during the dark phase was suppressed across treatments, while total distance increased at 20 and 50 µg/L, suggesting altered exploratory behaviour. At the molecular levels, 50 µg/L BPF upregulated bdnf,grin2aa, akt1, inhbab, and increased BDNF protein levels, while downregulating ntrk2a and sypa, indicating disrupted neurotrophic signalling, enhanced excitatory transmission, and altered synaptic regulation. In conclusion, embryonic BPF exposure disrupts neurotrophic and excitatory signalling, producing anxiety-like and hyperactive phenotypes. These findings identify BPF as a potential developmental neurotoxicant and highlight the need for long-term and mechanistic studies.

CHANGE I Integrating physiology and behaviour in a changing environment

Oxygen supply and demand in diel vertically migrating mesopelagic fishes (Myctophidae)

Seibel, Brad[1] (*seibel@usf.edu*) Oral Presentation
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Abstract: Oceans play a key role in global carbon budgets and cycles by absorbing ~30% of anthropogenic carbon dioxide. Understanding the mechanisms and drivers of the biological transport of carbon from the sea surface to the deep sea is essential for estimating the magnitude of net carbon flux into the oceans. Active flux is dominated by vertical migration of zooplankton and micronekton, which swim between productive epipelagic sunlit waters where they feed, and the mesopelagic "twilight" zone where they hide from visual predators during daylight hours. Myctophid fishes are the dominant mesopelagic fish family and are believed to contribute as much as 30% of the carbon flux from the surface to mesopelagic depths. At depth, diel migrators release carbon primarily through respiration, excretion and egestion. Estimates of the magnitude of this contribution are historically plagued by uncertainty in both fish abundance and physiology. The present project sought to alleviate that uncertainty using a unique suite of oceanographic and physiological tools. We report vertical distribution, abundance andmetabolic rates, as well as their oxygen- and temperature-dependence, in Myctophid fishes from the Gulf of California oxygen minimum zone.

CHANGE I Integrating physiology and behaviour in a changing environment

Biological responses of juvenile brook trout (Salvelinus fontinalis) to the interactive effects carbon dioxide and temperature

Bellinger, Imogen[1] (*imogen.bellinger@gmail.com*) Oral Presentation
Co-Author: Hasler, Caleb[1]; Raby, Graham[2]; Chasse, Samuel[1]
[1]The University of Winnipeg; [2] Trent University

Abstract: Environmental degradation is contributing to declines in freshwater biodiversity and ecosystem health. Two major threats to fish in these ecosystems are increasing temperature and rising carbon dioxide (CO2). While many studies examine the independent effects of CO2 or temperature on fish, few investigate their combined impacts, despite these stressors occurring simultaneously in nature. Furthermore, limited research has focused on freshwater ecosystems, which are highly variable and can experience both abrupt elevations in CO2, as well as concentrations substantially higher than those in marine environments. Here, we assessed the interactive effects of CO2 and temperature on juvenile brook trout (Salvelinus fontinalis). To do so, 1200 brook trout were divided into four treatments, including: control (~ 1000 uatm, 13°C), elevated temperature (~ 1000 uatm, 19°C), elevated CO2 (~5500 uatm, 13°C), and elevated CO2 and temperature (~ 5500 uatm, 19°C). Each

group was exposed to their treatment for 34-days and growth, metabolism, thermal tolerance, and behaviour were measured throughout. We found growth rates were highest in our control treatment and lowest in our elevated CO2 and temperature treatment. Standard metabolic rate increased in brook trout exposed to elevated temperature at the beginning of our exposure, but returned to baseline by the end of the exposure. This was not observed under elevated CO2. Thermal tolerance increased over the 34-day exposure under elevated temperature; however, in comparison, thermal tolerance was slightly reduced under elevated CO2 and temperature. Together, our results provide insight into what conditions will be suitable for brook trout as their environments change.

CHANGE I Integrating physiology and behaviour in a changing environment

CO2 induced spatial working memory impairment and recovery in medaka (Oryzias latipes)

Hryciw-Pang, Sage (*pang_sage@hotmail.com*) Oral Presentation
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Abstract: Fish use navigation strategies to perform fitness related tasks such as foraging, predator avoidance, habitat selection, and/or migration. These tasks rely upon the executive function of spatial working memory (SWM), which can be influenced by environmental changes. As atmospheric carbon dioxide (CO2) levels rise, freshwater ecosystems are left vulnerable to fluctuations in pH, which results in aquatic acidification. Previous research using the laboratory species medaka (Oryzias latipes) has resulted in changes in transcript abundance in various organs, including the brain, from increased CO2 exposure. Whether these changes influence SWM has not been explored and was the aim of this study. To investigate how CO2 exposure affects SWM in fish, 112 adult female and male medaka performed a total of four 1-hour trials in a free-movement pattern Y-maze over the span of 15 days. Trials included a baseline (day 0), CO2 exposure (day 5, 5615 ppm, pH = 6.3), recovery from CO2 exposure (day 10), and exposure to MK-801 (day 15, 0.75mg/L), a known working memory inhibitor. Changes in search patterns based on turn choices (left or right) were analyzed as overlapping tetragrams and compared to baseline trials to analyze the effects of treatment. Preliminary results suggest that medaka show a dominant turning choice for alternating search patterns and that MK-801 successfully disrupts the acquisition of prior turn choices by decreasing the number of alternations made. Acute CO2 exposure does not seem to influence search strategies in medaka. Understanding how aquatic acidification affects SWM in fish allows us to continue to predict how fish navigate in their environments.

CHANGE I Integrating physiology and behaviour in a changing environment

Changing temperatures: can cool water recovery offset high temperatures for partially armored threespine stickleback native to California?

Cooper, Cassidy[1] (*cjcooper@ucdavis.edu*) Oral Presentation
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Abstract: The Santa Clara River in southern California (USA) is a heavily modified, intermittent freshwater stream with significant seasonal and geographic thermal variation. Threespine stickleback (Gasterosteus aculeatus) is one of the most thermally-sensitive native species of conservation concern in the area, and understanding the role of thermal refugia provides useful insight for management in such heterogeneous environments. We measured the CTMAX of fish (n = 48) exposed to either two constant acclimation temperatures (25.6 and 26.7°C) or a cycling thermal regime that oscillated between 22.8 - 28.3°C (mean temperature = 25.6°C). Fish exposed to daily temperature cycles showed higher CTMAX when sampled at the warm part of the cycle. Acclimation to high temperatures led to decreased survival for fish, although exposure to cycling temperatures appeared to delay the onset of mortality. We also measured the temperature preference of fish (n = 20) acclimated to 18.6°C (an indefinitely sustainable temperature) using a dynamic two-chambered shuttle box system. Mean temperature preference was 18.0°C. Measured values fell between 10.7 - 24.1°C, and fish avoided temperatures >24.4°C, even when temperatures up to 30°C were available. Our data suggests that prolonged exposure to >25°C causes mortality. In thermally-challenging systems like the Santa Clara River, access to cool water refugia may underpin persistence for species living at or near their thermal limits. Future research is needed to further investigate the physiological impact of cool recovery temperatures as well as the extent to which thermally-impaired native species successfully utilize them in a natural context.

CHANGE I Integrating physiology and behaviour in a changing environment

Effects of livewell chilling on the physiology and behaviour of smallmouth bass

Thapar, Myra (*myrathapar@cmail.carleton.ca*) Oral Presentation
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Abstract: Livewell confinement is a common practice in catch-and-release fishing, especially in tournaments. However, this practice can expose fish to crowding and abrupt water changes. Many anglers add ice to their livewells to keep fish cool over the course of fishing. However, this subjects fish to “cold shock”, a phenomenon that occurs when water temperatures rapidly move away from usual conditions. Cold shock can occur due to natural or anthropogenic causes that influence both the physiology and behaviour of fish. If not managed carefully, it can cause harm to fish and influence their recovery after release. This study examined how livewell temperature and holding duration affect smallmouth bass (Micropterus dolomieu) at the molecular (gene expression) and behavioural

(temperature and depth preference post release) levels. Smallmouth bass were angled and held in livewells at various temperatures (15, 20, and 25°C) as well as various durations (1, 3, and 5 hours). Non-lethal gill samples were taken from fish prior to release. Short-term behaviour after release was monitored through the use of accelerometers that detect temperature and depth preference for 10 minutes post-release. A transcriptomics approach was used to assess changes in gene expression via a stress transcriptional profiling chip post treatment. By linking molecular responses with short-term post-release behaviour, this work furthers our understanding of the overall effects of cold shock and helps inform evidence-based guidelines for livewell temperature management in an effort to better fish health. This information helps guide best practices for fish handling during fishing tournaments and catch-and-release angling.

CHANGE I Integrating physiology and behaviour in a changing environment

Seasonal habitat selection and activity patterns of juvenile burbot (Lota lota) in a laboratory setting

Anderson, Lars (*larsbanderson@gmail.com*) Poster Presentation
Co-Author: Leonard, Jill
Northern Michigan University

Abstract: Burbot (Lota lota) are the sole freshwater representative of the Order Gadiformes, which includes marine cod species, and have a circumpolar distribution north of 40o . Despite the ubiquity of burbot, little is known about the habitat selection and activity patterns of juveniles and how these behaviors shift across distinct seasons. Quantifying these behaviors is essential for understanding the energetic trade-offs and habitat requirements that underpin early-life survival, particularly as northern systems experience rapid winter warming. We investigated juvenile burbot habitat use and activity using radio frequency identification (RFID) telemetry within a controlled artificial stream system containing both pool and moderate-flow riffle sections. We simulated seasons by manipulating water temperature and photoperiod to reflect winter and summer conditions. Within each seasonal treatment, we compared responses to varied structural habitats (wood versus rock complexes). Under simulated winter conditions, juveniles favored riffle habitats over pools with occupancy jumping from 49.1% in summer to 62% in winter. Habitat structure further shaped occupancy patterns: wood treatments elicited significantly greater site fidelity and prolonged residency compared to rock substrates. Activity varied with both habitat type and season, but overall increased more than 3.5-fold from summer to winter. Notably, this heightened winter activity contrasts with the reduced activity observed in many other freshwater species during cold periods. Understanding the seasonal habitat selection and activity patterns of juvenile burbot is increasingly important as climate-driven shifts in temperature regimes across northern systems may have cascading consequences for juvenile survival and long-term population stability in this cold-adapted species.

CHANGE I Integrating physiology and behaviour in a changing environment

Seasonal patterns of brain size plasticity in freshwater fish

Francispillai, Erin (*efranc06@uoguelph.ca*) Poster Presentation
Co-Author: Turko, Andy and Laberge, Fred
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Abstract: Phenotypic plasticity allows organisms to shift physiological, behavioural, and/or morphological traits to respond to environmental change within their lifetime. In fishes, one highly plastic organ is the brain, which exhibits high rates of neurogenesis and can show reversible plasticity into adulthood. However, data are available only for a handful of species, so the scope for brain plasticity among species and the causal mechanisms for this plasticity are unknown. To investigate seasonal patterns of brain size plasticity among fishes, we measured brain mass of 20 species native to southern Ontario in summer and winter of 2024 and 2025. We discovered divergent plastic responses in fish brain size, with some species showing the largest brain size in the summer, others in the winter, and others showing no plasticity. Thus, brain size plasticity may be species-specific. Evaluating if species can respond to current levels of environmental change through trait plasticity may provide insight into their ability to respond to projected change and may be key in determining vulnerability. Finally, all captured fish were preserved in formalin solution; however, prolonged formalin preservation may reduce brain mass measurements, introducing an effect of time since capture. Thus, we also include preliminary data on the effect of formalin preservation on brain mass over time, suggesting a plateau in effects that should be considered for physiological research across taxa.

CHANGE I Integrating physiology and behaviour in a changing environment

Acute resilience, chronic costs: metabolic responses to warming and hypoxia in the sedentary Lusitanian toadfish, Halobatrachus didactylus

Guerreiro, Pedro M[1] (*pmgg@ualg.pt*) Poster Presentation
Co-Author: Molina, Juan M[2]; Kunzmann, Andreas[3]; Costa, Rita A.[1]; Alves, Alexandra[1]; and Modesto, Teresa[1]
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Abstract: Coastal fishes can adapt to water warming and hypoxia, however acute tolerance does not necessarily predict longer-term performance and survival. This may be especially important in sedentary, site-faithful species with limited escape to escape increasingly unfavourable habitats. We assessed the climate-related stress responses in the Lusitanian toadfish, Halobatrachus didactylus, a benthic estuarine fish from the Northeast Atlantic to simulated water warming and hypoxia.
We determined standard, routine, and maximum metabolic rates, aerobic scope, critical thermal maximum (CTmax), and critical oxygen thresholds (Pcrit) on a set of XX individuals. On a different set of individuals we simulated 30 days of seasonal scenarios combining low

and high temperature with normoxia or hypoxia, followed by integrated metabolic, hematological, biochemical, and multivariate analyses.
Acute trials showed high short-term resilience: H. didactylus had an exceptionally low standard metabolic rate, routine metabolic rate, high CTmax (34.82 ± 0.66 °C), and strong hypoxia tolerance (Pcrit 0.59–1.97 mg O2 L^{-1}), although smaller individuals were more sensitive. After 30 days, however, warming more than doubled standard and routine metabolic rates, while warm hypoxia reduced metabolic output relative to warm normoxia, consistent with metabolic depression under compounded stressors. This treatment also showed shifts in glucose, liver mass, red blood cell count, and hematocrit, identifying warm, oxygen-poor water as the most physiologically costly scenario for this species. Together, these results show that high acute tolerance does not guarantee resilience to climate change. In sedentary fishes, survival may depend less on surviving extremes than on maintaining energetic balance, oxygen transport capacity, and physiological homeostasis in increasingly warm, oxygen-poor coastal habitats.

CHANGE I Integrating physiology and behaviour in a changing environment

Heat hardening in grey mullets: Physiological responses of juvenile Chelon labrosus and Chelon aurata under simulated short-term marine heatwaves

Guerreiro, Pedro M (*pmgg@ualg.pt*) Poster Presentation
Co-Authors: Amaral, Inês[1]; Costa, Rita A.[1]; Zamora-López, Antonio[2]; Zimmermann, Wim[3] and Gregorio, Silvia F.[1]
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Abstract: Marine heatwaves are increasing in frequency and intensity, posing major challenges for fishes inhabiting shallow coastal systems. We examined the effects of short-term warming on thermal tolerance and metabolic performance in juvenile grey mullets, Chelon labrosus and Chelon aurata, two abundant species forming sympatric schools in Ria Formosa (southern Portugal). Fish were exposed to simulated heatwave treatments of 23, 27, or 33°C, after acclimation at 17°C, and were sampled either at peak temperature or after 48h and 1-wk recovery at 17°C. Critical thermal maximum (CTmax), static oxygen consumption (MO2), and intermittent respirometry traits were measured.
CTmax increased significantly with warming in both treatment modes, demonstrating rapid heat hardening. Fish exposed to 27 and 33°C showed higher CTmax than control fish, and this elevated tolerance persisted after recovery. A linear model further indicated that C. labrosus had slightly higher CTmax than C. aurata. Oxygen consumption also increased with temperature, with the strongest responses at 33°C. Intermittent respirometry showed that standard metabolic rate rose markedly with warming, especially in heatwave-exposed fish, while maximum metabolic rate increased mainly at the highest temperature. In contrast, aerobic scope showed no clear thermal optimum or decline across treatments. In a complementary temperature-ramp experiment, routine metabolic rate increased non-linearly with temperature and Q10 averaged 2.28, confirming a strong thermal dependence of metabolism.
These results show that juvenile mullets possess substantial short-term thermal plasticity, but this increased heat tolerance is accompanied by higher metabolic costs under extreme

warming, but differential responses may predict school composition at thermal landscapes. Further analysis on plasma and tissues will characterize the mobilization of energetic substrates and the cellular responses to thermal and oxidative stress.

CHANGE I Integrating physiology and behaviour in a changing environment

ShoalBase: Global community database of fish social behaviour

Killen, Shaun[1] (*shaun.killen@glasgow.ac.uk*) Poster Presentation
Co-Authors: Cortese, Daphne[2]; Munson, Amelia[3]; Kochhann, Daiani[4]; Pineda, Mar[1] and Tiddy, Izzy C.[1]
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Abstract: Understanding social behaviour across fishes remains surprisingly difficult despite fishes representing more than half of all vertebrate diversity and exhibiting an extraordinary range of social systems, from solitary living to highly coordinated schooling. Existing knowledge is fragmented across literature sources, personal observations, and community knowledge, making broad comparative synthesis challenging. To address this gap, we have created ShoalBase, an open-access and community-driven database for collating records and observations of fish social behaviour across species and contexts. ShoalBase integrates contributions from researchers, aquarists, field observers, citizen scientists, fishers, and published literature, allowing users to submit observations through a simple structured interface that records species identity, life stage, group size, social category, habitat, environmental context, evidence type, and source information. Observations can derive from wild, captive, or experimental settings, enabling comparison across ecological and observational contexts while explicitly documenting evidence quality and potential observer bias. The platform is designed not only as a repository but also as an exploratory tool, with interactive dashboards that summarize taxonomic, geographic, and behavioural coverage, and a phylogenetic interface that allows users to visualize social traits across fish lineages. Early submissions already reveal strong biases toward adult life stages, conspicuous species, and certain geographic regions, highlighting major knowledge gaps such as the near absence of larval-stage social data. By creating a scalable and community-driven resource, ShoalBase aims to accelerate comparative research on fish social evolution, identify priority areas for future study, and provide a foundation for linking social behaviour with ecology, physiology, conservation, and environmental change.

CHANGE I Integrating physiology and behaviour in a changing environment

Methane exposure alters the transcriptome and behaviors of developing zebrafish (Danio rerio)

Steffler, Morgan (*msteffle@ualberta.ca*) Poster Presentation
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Abstract: Nearly half of global methane emissions originate from aquatic systems, including both natural and anthropogenic environments. In end pit lakes, abundant methanogens elevate dissolved methane concentrations, warranting consideration in bioremediation efforts. In both urban and rural environments, methane accumulation in shallow aquifers that supply lakes, rivers, and drinking water sources is also a concern. Although recognized as a flammable, explosive, and asphyxiant gas, methane is not currently classified as a health hazard due to limited toxicological research. An absence of evidence does not eliminate risk, making investigation into its sublethal consequences essential. In our study, we exposed 0.5-4 dpf zebrafish to a range of environmentally relevant methane concentrations, and used RNA-seq and behavioural analysis to characterize any exposure effect(s). Exposures altered ~200 genes related to metabolic pathways and ~500 genes associated with cell membranes. With behaviour, an unexpected non-monotonic response was noted, where fish exposed to low methane concentrations were less active than those exposed to medium but not higher doses. This pattern was apparent in distance travelled, average velocity, maximum velocity, cumulative time spent moving, and latency to first movement. Our findings suggest that early-life methane exposures may cause a suite of adverse outcomes that reduce survival and alter ecological interactions.

CHANGE I Integrating physiology and behaviour in a changing environment

Coping with cloudy streams: Mechanisms of suspended sediment tolerance in fishes

Young-Veenstra, Sarah (*syoung14@uoguelph.ca*) Poster Presentation
Co-Authors: Turko, Andy
University of Guelph

Abstract: Suspended sediment (SS) occurs naturally in aquatic systems, but anthropogenic activities often increase SS beyond natural levels, contributing to declines in fish abundance and diversity. The mechanisms underlying SS-induced harm and interspecific differences in sensitivity are poorly understood. One possibility is that SS impairs respiration by clogging gill lamellae via sediment or mucus accumulation (“clogging” hypothesis) and/or physically abrading the gill epithelium (“damage” hypothesis). Alternatively, SS may induce maladaptive behavioural responses such as erratic swimming (“panic” hypothesis) or disrupt visually mediated behaviours including foraging and predator avoidance (“sensory” hypothesis). Despite their prominence, there are few empirical tests of these ideas. To test these ideas, I will first establish a continuum of SS tolerance across ~20 species of North American stream fishes by gradually increasing SS

concentrations and identifying tolerance thresholds. I will test the clogging hypothesis by measuring branchial resistance and examining gill traits associated with clogging susceptibility. Next, I will expose species to chronic SS followed by recovery in clear water. Throughout exposure and recovery, I will test the (i) damage hypothesis by quantifying gill morphologies indicative of beneficial plasticity and tissue damage; (ii) panic hypothesis by measuring changes in diel swimming behaviour; and (iii) sensory hypothesis by quantifying prey detection and capture rates in the presence and absence of a chemical predator cue. Overall, this research will be among the first to investigate mechanism(s) behind SS-induced harm and tolerance, which may help predict changes in biodiversity and inform conservation decisions.

TOXICOLOGY Fish talks on phys tox

Fish talks on phys tox

Sarah Alderman and Todd Gillis

Abstract: Fish physiologists are critical players in the field of aquatic toxicology, providing mechanistic insight into contemporary challenges that impact the health and fitness of individual fish and, by extension, entire ecosystems. The list of established and emerging chemicals of concern is long and growing. From embryos to food webs, PAH to PFAS, the goal of this session is to highlight research across a breadth of study systems, pollutants, and methodologies, emphasizing sublethal effects and opportunities for knowledge translation.

The Impacts of Dietary PFOA on the Reproductive Behavior of Male Zebrafish (Danio rerio)

Khursigara, Alexis (*khursigaraa@montclair.edu*) Oral Presentation
Montclair State University

Abstract: Nicknamed “forever chemicals”, per- and polyfluoroalkyl substances (PFAS) are manmade chemicals that are ubiquitous in the aquatic environment. Despite its production and use being heavily phased out, one of the most prevalent PFAS in the environment is perfluorooctanoic acid (PFOA). Recent research has highlighted the suite of injuries that PFOA can induce in aquatic organisms like fish, including changes in behavior, growth, development, reproductive success, and endocrine function. Despite the growing body of research, the effects of chronic dietary exposure of adult fishes have been understudied. To address this knowledge gap, the effects of three dietary concentrations of PFOA were assessed on male zebrafish (Danio rerio). After 1, 2, and 4 weeks of exposure, length and mass were taken after a 10-minute mating trial. Videos of the mating trials were assessed for distance moved, time spent moving, average velocity, average distance to the female, time in contact with the female, and various mating behaviors. Preliminary results indicate an effect of PFOA on growth, distance traveled, and velocity. Since males must be active during mating and often chase females to get them to release eggs, changes in these parameters can have significant implications for reproductive success for male fish.

TOXICOLOGY Fish talks on phys tox

Assessing Lithium Toxicity Across a Salinity Gradient in Coastal Marine Fishes

Esbaugh, Andrew (*a.esbaugh@austin.utexas.edu*) Poster Presentation
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Abstract: Lithium use has been steadily increasing with the proliferation of rechargeable electrical devices worldwide, which has led to an increase in aquatic lithium pollution. In freshwater, lithium is thought to enter the animal through sodium uptake pathways. Consistent with this hypothesis, sodium is protective against lithium toxicity in freshwater fish. However, marine fishes drink seawater and actively absorb the majority of imbibed ions, which may result in the loss of sodium protection in hyperosmotic environments. The current study explored lithium toxicity across a salinity gradient in two coastal fish species, the sheepshead minnow (SHM) and red drum, using a combination of developmental and behavioral approaches. Contrary to conventional understanding, the lowest tested salinity had the highest developmental EC50 for both species – although for both species the observed toxicity was well in excess of typical environmental scenarios. Interestingly, developmental toxicity coincided with a significant decline in heart rate and an increase in pericardial edema. A series of behavioral open field tests following 96-h of exposure in larval SHM (21 dpf) also revealed significant hypoactivity with exposed fish having significantly reduced percent active time, distance travelled, and average speed than controls. Notably, behavioral impairments were significantly more sensitive than developmental toxicity endpoints. Overall, these data suggest that caution should be exercised when extending freshwater models of lithium toxicity to coastal environments, and that behavioral toxicity may provide a more sensitive indicator of lithium toxicity in fishes than traditional approaches.

TOXICOLOGY Fish talks on phys tox

Timing is everything: Critical windows of road salt toxicity in early life stage Pacific salmonids

Kilgour, Clare L. (*ckilgour@zoology.ubc.ca*) Oral Presentation
Co-Authors: Hall, Jack; Winter, Carley E.; Whelan, Piper; Brauner, Colin J.; Wood, Chris and Schulte, Patricia
The University of British Columbia

Abstract: In the Vancouver Lower Mainland, road salt (NaCl) washes into local freshwater streams during the fall and winter, resulting in acute pulses of increased salinity. During this same period, Pacific salmon return to these freshwater streams to spawn, depositing their eggs in gravel redds where embryos develop throughout the winter. In previous investigations, we have seen that these road salt pulses can be toxic to early life stage rainbow trout, coho salmon and chum salmon at concentrations regularly observed in streams. However, both the timing and frequency of the exposure are important factors that influence the ultimate toxicity of road salt pulses. Notably, the first day of development appears particularly sensitive to road salt exposure, indicating a potential critical window

of vulnerability. To further investigate this critical window and better understand the mechanisms of road salt toxicity, we exposed rainbow trout embryos to 6-hour road salt pulses at one of five timepoints during the first 30 hours of development. The exposed embryos were sampled for whole body ions, preserved for developmental staging, and tested for endpoints related to egg hardening (size and rupture force) (NSERC Alliance).

TOXICOLOGY Fish talks on phys tox

Impacts of road salt exposure on water hardening in coho salmon eggs (Oncorhynchus kisutch)

Hall, Jack (*Jackhall@student.ubc.ca*) Oral Presentation
Co-Authors: McCarthy, Desmond; Kilgour, Clare L.; Winter, Carley E.; Brauner, Colin J.; Wood, Chris and Schulte, Patricia
The University of British Columbia

Abstract: Freshwater salinization events due to road salt runoff are both widespread and severe in British Columbia's lower mainland. Recent research has indicated coho salmon (Oncorhynchus kisutch) eggs may experience high mortality if exposed to road salt early in development, and that this mortality may be ameliorated through the addition of waterborne calcium. It was hypothesized that mortality may be caused by salt interrupting the egg's water hardening process; this seemed a likely mechanism, as water hardening occurs early in development, has a protective role for the embryo, and is initiated through the intracellular release of calcium ions. To investigate, coho eggs were exposed to one of six treatments for the first 24 hours after fertilization (2400 mg/L Cl, 4800 mg/L Cl, 2400 mg/L Cl + 200 mg/L CaCO3, 4800 mg/L Cl + 200 mg/L CaCO3, 200 mg/L CaCO3, Freshwater Control). The force required to rupture the eggs was then repeatedly tested using a motorized force meter. By 48 hours after fertilization, both the 2400 mg/L Cl and 4800 mg/L Cl treatment groups required significantly greater force to rupture than the control group. However the hardening process was delayed in the 4800 mg/L group; from 0-24 hours post fertilization they had not experienced any hardening at all. The addition of calcium did not have a significant effect in any treatment, suggesting that changes to the egg hardening process are unlikely to be the primary mechanism of mortality (NSERC Alliance).

TOXICOLOGY Fish talks on phys tox

Effects of Chronic Nitrite Exposure on Reproduction and Maternal Transfer in a Live-Bearing Fish Model (Xiphophorus couchianus)

Parkman, Trinity (*vcm60@txstate.edu*) Oral Presentation
Co-Author: Huertas, Mar and Lu, Yuan
Texas State University

Abstract: Fertilizer runoff from agricultural practices can lead to high nitrite concentrations in aquatic environments and drinking water. Nitrite has been well studied for its acute toxic effects; however, its chronic effects and effects on reproduction and development remain understudied. We hypothesized that nitrite would alter or inhibit maternal transfer – a key mechanism to provide temporary immunity and proper development – of steroid and immune factors during gestation. To investigate this, we exposed a live-bearing fish model, Xiphophorus couchianus, to nitrite concentrations of 0 mM, 0.01 mM, 0.1 mM, and 1 mM. Following their 30-day gestation, females were sacrificed to obtain embryonic sacs for staging, quantification, and histological assessment. Head kidneys, oocytes, and pseudoplacental tissue were preserved for RNA extraction and TaqSec transcriptomics. Using counts of each identified stage, we determined that chronic nitrite exposure significantly suppresses fertilization and embryonic development. Compared to the controls, the histology of the 1 mM-treated females showed increased blood flow and methemoglobin presence, an increased prevalence of macrophages, melanomacrophages, and lymphocytes, and increased atresia in pre-vitellogenic oocytes. RNA sequencing showed changes in immune and steroidogenic gene expression with increasing nitrite exposure. Overall, these results suggest that chronic nitrite exposure stimulates immune responses in the head kidney and ovaries and represses oogenesis and embryogenesis by interfering with steroidogenesis. These findings could have further implications for how prolonged nitrite exposure affects pregnancy in mammals, particularly in human women. Therefore, it is advised that nitrite levels should be routinely monitored in aquaculture and drinking water to reduce exposure risk.

TOXICOLOGY Fish talks on phys tox

The common pesticide azoxystrobin is toxic to the fish heart

Ruhr, Ilan[1] (*ilanruhr@gmail.com*) Oral Presentation
Co-Author: Shiels, Holly[2]
[1] University of Salford; [2] The University of Manchester

Abstract: By 2050, the Earth's human population will grow by 25% to 10 billion people, requiring a 50% increase in worldwide agricultural production. Consequently, pesticide use will inexorably grow, but current usage patterns pose an existential threat to wildlife. In this study, we investigated the sublethal cardiac effects a globally used fungicide, azoxystrobin, has in goldfish (Carassiusauratus). Azoxystrobin enters aquatic ecosystems, via agricultural runoff/drift, and targets mitochondrial respiration, by binding to complex III of the mitochondrial electron-transport system (ETS) and disrupting ATP production via oxidative phosphorylation (OXPHOS). We have gathered the first-ever evidence showing

that azoxystrobin impairs contractile function in ventricular cardiomyocytes (heart cells) at concentrations (<3.3 µg/L) lower than regulatory safe environmental concentrations. Specifically, we used epifluorescent microscopy to determine that azoxystrobin reduces cardiomyocyte shortening, by diminishing intracellular calcium transients, the chief regulators of contractile force. Because ATP production is a key component of cardiomyocyte contractility, we used microrespirometry to find that azoxystrobin suppresses OXPHOS and elevates the generation of harmful reactive oxygen species in cardiac mitochondria. Goldfish are considered one of the most anoxia-tolerant vertebrates, yet our data show that their cardiomyocytes are profoundly sensitive to azoxystrobin. Considering that azoxystrobin mimics hypoxic conditions, by preventing oxygen from acting as a final electron acceptor on the ETS, hypoxia-sensitive fish in the wild are likely to be more affected by azoxystrobin exposure than goldfish. By characterising the sublethal cardiovascular risks of agricultural pesticides, we can adapt current usage patterns to improve the survivability of wild aquatic animals, while boosting agricultural production.

TOXICOLOGY Fish talks on phys tox

Alterations in transepithelial potential and ionoregulation in goldfish following exposure to differentially degraded microplastics in two different water chemistries

Zink, Lauren[1] *lauren.zink@zoology.ubc.ca* Oral Presentation
Co-Authors: Griffiths, Jaden[2]; Winter, Carley E. [1]; Morris, Carolyn[1]; Wiseman, Steve[2] and Wood, Chris[1]
[1] The University of British Columbia; [2] University of Lethbridge

Abstract: Microplastics (MPs) are constantly degrading while moving through aquatic systems as a result of mechanical abrasion, thermal fluctuations, UV light, and chemical exposure. As such, fish may experience pulse exposures to differentially degraded MPs. MPs have been observed to interact with the fish gill during extended exposures (hours – days), causing increased mucus production, damage to secondary lamellae, and oxidative stress, suggesting disturbances to ionoregulation but there have been few direct measurements. We used transepithelial potential (TEP) across the gills as an indicator of gill functionality. Under control conditions, TEP (the diffusion potential between the blood plasma and external water) is negative. Virgin (non-degraded) MPs along with mechanically, UV, and thermally degraded plastics immediately depolarized the gills (less negative TEP), whereas chemically degraded MPs resulted in no change to TEP. We then assessed net flux of major cations (calcium, sodium, potassium, and magnesium), nitrogenous wastes (ammonia, urea), and chloride over a 6-hour exposure to virgin MPs or chemically degraded MPs. We isolated gill and digestive tissue to explore whether MP uptake is significant over the course of 6 hours. Finally, as water chemistry can alter both TEP and the behaviour of MPs, we mirrored these experiments across two different water chemistries that varied in hardness, alkalinity, and pH (NSERC).

TOXICOLOGY Fish talks on phys tox

Signals in the stream: Field-to-lab evidence of molecular and biochemical stress in fish exposed to wastewater-borne drugs of abuse

Craig, Paul[1] (*paul.craig@uwaterloo.ca*) Oral Presentation
Co-Author: Rowland, Zoe[1]; LeBlonc, Melissa[1]; Nussbaum, Benjamin[1]; Fuzzen, Meghan[1]; Tetrault, Gerald[2] and Servos, Mark[1]
[1]University of Waterloo, [2]Environment and Climate Change Canada

Abstract: Wastewater treatment plant (WWTP) effluent represents a continuous source of complex contaminant mixtures to freshwater ecosystems, including pharmaceuticals and drugs of abuse that persist at environmentally relevant concentrations. We evaluated the biological impacts of effluent exposure in the Grand River (Brantford, Ontario) by integrating field surveys of endemic darters with laboratory-based exposures in a model species. Six sites spanning upstream reference locations and downstream reaches of the Brantford WWTP outfall were surveyed, focusing on greenside darter (Etheostoma blennioides) and blackside darter (Percina maculata). Across species, we observed a consistent pattern of elevated oxidative stress biomarkers downstream of the effluent discharge. Antioxidant enzyme activities and transcript levels were generally increased in downstream fish, accompanied by higher indices of oxidative damage, suggesting chronic reactive oxygen species (ROS) induction in resident populations exposed to effluent-associated contaminant mixtures. To mechanistically link exposure to biological response, we conducted controlled early-life stage screens in fathead minnows (Pimephales promelas). Larval exposures to environmentally relevant concentrations of selected psychoactive compounds and effluent-informed mixtures altered expression of genes associated with oxidative stress, neurotransmission, and growth regulation, indicating disruption of conserved molecular pathways. Together, these results provide convergent field and laboratory evidence that WWTP effluent in the Grand River is associated with measurable neurophysiological and oxidative stress responses in fish. By comparing wild populations with controlled exposures, this work advances our understanding of how complex contaminant mixtures translate into biological effects, strengthening risk assessment frameworks for effluent-impacted watersheds.

TOXICOLOGY Fish talks on phys tox

Thyroid receptor activity reporter: modelling the fundamental mechanisms of thyroid disrupting pollutants in a changing climate

Simm, Hayden (*simmh@mcmaster.ca*) Oral Presentation
Co-Author: Little, Alexander
McMaster University

Abstract: Climate change is intensifying themagnitudeofabioticstress imposed on animals by their environment.Simultaneously,novel stressorsincludingendocrinedisrupting pollutants(EDPs)areincreasinglyubiquitous in the environment.Thiscombinationisconcerningbecauseendocrinesystems playa central rolein mediating physiologicalremodellingto overcomeenvironmental stress.Particularly,the

thyroid axiscoordinatesmetabolic responses to temperature (thermal plasticity)in fish, and thyroid disrupting pollutants(TDPs)are a prolific class of EDPs.Thus,the increasing prevalence ofTDPs mayimpair thyroid signalling andcompromisethecapacity tocompensate for thermal variability associated with climate change.To understand these potential interactions, wegeneratednovelTRactivity reporterlines ofzebrafish (Danio rerio), which allowus to quantifyTRactivity using a fluorescentreporterand thus measure endocrine disruption at the most fundamental level of action.Bytargetingthe action of TRs we will(i)characterizetemporalandtissue specificthyroid-mediatedresponsesto thermal stressduring development,and(ii)contrastTR activitypatterns whenTDPsare introducedduringthe mostcritical windowsin dynamic thermal environmentsto assesseffect on plastic remodelling.This integrative approach willprovidemechanistic insight into how environmental stress and pollutant exposure interact at multiple levels of biological organization. Byidentifyingsub-lethal thresholds of thyroid disruption in ecologically relevant conditions, this research will improve our ability to predictand modelthe consequences of contaminant exposure in the face of climate change.

TOXICOLOGY Fish talks on phys tox

Environmental fate meets fish function: rethinking risk of sea lice chemotherapeutants in coastal salmon habitat

Kennedy, Chris[1] (*ckennedy@sfu.ca*) Oral Presentation
Co-Authors: Baillon, Lucie [2]; Langlois, Valerie [2] and Lin, Feng [1]
[1] Simon Fraser University; [2] Institu National de la Recherche Scientifique

Abstract: Sea lice chemotherapeutants used in open-net salmon aquaculture are deliberately released into coastal waters that juvenile pink salmon (Oncorhynchus gorbuscha) must traverse during out-migration. To move beyond traditional lethality metrics, we linked environmental fate to organismal performance in seawater-acclimated fry. Water–sediment microcosms revealed distinct exposure pathways: azamethiphos (AZ) and hydrogen peroxide (HP) remained largely in the water column and dissipated relatively quickly, whereas cypermethrin (CP), deltamethrin (DM), and emamectin benzoate (EB) rapidly partitioned to sediments and persisted for weeks to months (sediment half-lives up to ~557 d for CP). These dynamics define realistic exposure scenarios: short pelagic pulses for AZ and HP, but prolonged benthic exposure potential for pyrethroids and EB in nearshore habitats used by migrating salmon. Acute toxicity in fry followed the gradient HP < EB < AZ < CP < DM, with DM most potent. However, mortality substantially underestimated risk. At concentrations well below LC_{50} values, AZ, CP, and DM reduced olfactory responsiveness to food cues, and CP impaired olfaction following sediment exposure. Critical swimming performance declined by up to 80% after brief sublethal exposures, and avoidance behaviour was limited or absent for the most potent compounds. Together, these findings demonstrate that partitioning governs where these chemicals go, persistence governs how long they remain bioavailable, and functional endpoints reveal ecological consequences invisible to standard acute tests. Integrating

fate with performance provides a more realistic framework for evaluating aquaculture chemical risk to migrating Pacific salmon.

TOXICOLOGY Fish talks on phys tox

Acute impacts of wildfire ash exposure on larval Pacific coho salmon (Oncorhynchus kisutch) physiology and survivorship

Munro, Cole (*colemun@student.ubc.ca*) Oral Presentation
Co-Author: Frommel, Andrea and Wood, Chris
The University of British Columbia

Abstract: As a result of climate change, wildfires are occurring at higher frequencies and intensities than historically observed. This mismatch between current and historic disturbance regimes in coastal environments has resulted in unprecedented conditions within post-wildfire environments, primarily through the creation of ash and charred debris. Wildfire ash has been shown to present numerous physiological challenges when entering freshwater environments, ranging from exposure to toxicants such as polycyclic aromatic hydrocarbons (PAHs), mobilization of metals and dissolved organic carbon (DOC), and elevated pH and alkalinity that can last for years. However, there is currently little literature published on the impacts of ash exposure on the early life stages of salmon. Coho salmon (Oncorhynchus kisutch) are a Pacific salmon species whose early life stages commonly overwinter in isolated off-channel habitats, and as a result are potentially vulnerable to prolonged exposure to wildfire sediments. In this study, larval coho were exposed to varying concentrations of wildfire ash for 72 hours, ranging from 0.5g/L to 1.5g/L, to examine the acute impacts of ash exposure. Whole-body ammonia and urea were assayed alongside lipid content to determine the implications for ammonia metabolism, condition and short-term survivorship. Wildfires are seldom considered when it comes to salmon conservation due to their historic scarcity in coastal environments, and it is crucial the knowledge gap on their influences on early salmon life stages is understood to properly inform conservation management in a changing climatic landscape.

TOXICOLOGY Fish talks on phys tox

Cardiorespiratory alterations and arrythmias after acute exposure to water-accommodated fraction from crude oil in juveniles of two North American fish species

Weber, Lynn[1] (*lynn.weber@usask.ca*) Oral Presentation
Co-Author: April, Dan[1]; Taylor, Amy[1]; Palace, Vince[2]; Tomy, Gregg[3]; Peters, Lisa[2] and Janz, David
[1]University of Saskatchewan, [2]IISD – Experimental Lakes Area, [3]University of Manitoba

Abstract: Exposure of aquatic species to crude oil spills remains a continued threat in both marine and freshwater environments. While most research to date has focused on developmental effects, including often lethal cardiac deformities, direct functional effects and consequences to juvenile or adult life stages are poorly characterized. The aim of this

study was to compare acute cardiorespiratory changes in juvenile rainbow trout (Oncorhynchus mykiss) and fathead minnow (Pimephales promelas) after acute aqueous exposure to water-accommodated fractions (WAF) from crude oil. After 48 hr aqueous exposure to sublethal 30% WAF, trout had decreased cardiac output, depressed stroke volume, but no significant effect on heart rate compared to control trout. However, ECG analyses revealed significantly longer PR and QT intervals, accompanied by >50% incidence of arrythmias in exposed trout. In contrast, fathead minnows exposed to 30% WAF for 7 days showed increased cardiac output due to increased heart rate compared to control fish, but also showed WAF-induced arrythmia. Moreover, fathead minnows exposed to 10% or 30% WAF had impaired swim endurance (Ucrit) without a significant change in active or standard metabolic rate compared to control fish. Taken together, this confirms that acute aqueous exposure at the juvenile stage to sublethal crude oil impairs cardiorespiratory fitness in two important North American fish species, albeit with differing underlying responses. Moreover, cardiac arrhythmias, primarily atrioventricular block, are a common response in both species.

TOXICOLOGY Fish talks on phys tox

The response of Pacific salmon to diluted bitumen, working towards a lifecycle perspective

Gillis, Todd[1] (*tgillis@uoguelph.ca*) Oral Presentation
Co-Author: Kennedy, Chris[2]; Eliason, Erika [3]; Patterson, David [3]; Farrell, Tony [4] and Alderman, Sarah[1]
[1] University of Guelph; [2] Simon Fraser University, Canada; [3]Fisheries and Oceans Canada; [4]The University of British Columbia,

Abstract: The Trans Mountain pipeline carries diluted bitumen (dilbit), a type of crude oil, through the Fraser River watershed, one of the largest salmon producing systems in the world. Since 2014 our research team has been building a knowledge base on the consequences of environmentally-relevant dilbit exposures on the multiple life stages of salmon that exist within the watershed. This includes examining impacts on performance metrics, developmental effects, persistence/latency of toxic responses, life stage specific sensitivities, and the confounding effect of temperature on dilbit toxicity. Experiments with developing embryos demonstrate increased mortalities and latent mortality with exposure, that the response is affected by the timing of exposure during development and that a 3oC temperature increase amplifies the consequence of exposure. We have also shown that dilbit exposure of juvenile fish causes cardiac fibrosis, a reduction in aerobic capacity, and an increase in plasma biomarkers of neural-muscular damage. Currently we are working to determine how dilbit exposure affects the cardiorespiratory function of returning adult sockeye and coho, and their progeny, by measuring maximal metabolic rate (adults) and hypoxia induced loss of equilibrium (F1). By grounding our research in realistic exposure scenarios that connect dilbit exposure with sublethal endpoints relevant to the unique life history of Pacific salmon, we are generating data that can be used to help predict the

consequence of a dilbit leak on the vital rates of native fish populations. This work is supported by the Department of Fisheries and Oceans, Canada.

TOXICOLOGY Fish talks on phys tox

Phenanthrene is a ubiquitous cardiac toxicant with exacerbated impacts on fish

Shiels, Holly (*holly.shiels@manchester.ac.uk*) Oral Presentation
University of Manchester

Abstract: Polycyclic aromatic hydrocarbons (PAHs) are environmental contaminants generated mainly by industrial activity and the incomplete burning of organic materials. Because they persist in air, water, and soil, they disperse widely and create sustained exposure risks for humans, wildlife, and ecosystems. Among these compounds, the three-ring PAH phenanthrene has attracted attention due to its distinct cardiotoxic effects. This talk will provide an overview of the effects of phenanthrene, and its derivatives, on cardiac function with a focus on fish.I will describe how phenanthrene and its derivatives disrupt cardiac electrophysiology by inhibiting repolarizing potassium currents (e.g., IK) and suppressing depolarizing sodium and calcium currents (e.g., INa and ICa), thereby increasing susceptibility to arrhythmias across a range of fish species. The impact of these pollutants on cardiac contractility will also be described through data on intracellular Ca^{2+} transients and contractility of isolated muscle.Mechanisms underlying the greater phenanthrene-sensitivity of fish hearts compared with other vertebrates' hearts will be explored.The lecture will end by briefly highlighting why fossil fuel pollution is also concerning for the hearts of humans inhaling polluted air.

TOXICOLOGY Fish talks on phys tox

Caffeine as an emerging contaminant: behavioral and physiological effects of acute exposure in zebrafish

Chaves Morera, Victoria Poster Presentation
Co-Authors: Vidaurre, Naidely; Villegas, Eliana; Zamora Villalobos, Esteban; Akerman Sánchez, Johanna Galit and Castro Murillo, Maripaz
University of Costa Rica

Abstract: Caffeine is the most consumed psychostimulant substance globally, and is frequently detected in aquatic environments due to wastewater discharge. As an emerging contaminant, its sublethal effects on fish physiology and behavior remain insufficiently characterized. This study evaluated the physiological and behavioral responses of zebrafish (Danio rerio) following acute exposure (7 days) to caffeine (0.0025 µg/L). Two tanks were established: a control group (n = 12) and a treatment group (n = 12) each containing individuals of both sexes. Individuals were assessed for locomotion and phototaxis behavior, ventilatory frequency, and metabolic rate using intermittent-flow respirometry. Caffeine exposure significantly increased locomotor activity ($p < 0.001$), dark-area preference ($p = 0.021$) and ventilatory frequency ($p = 0.044$). No statistically

significant effect on metabolic rate was detected; however, mean oxygen consumption values were higher in the caffeine-exposed group. These findings suggest that environmentally relevant concentrations of caffeine alter both behavior and physiological variables, potentially increasing energetic demand. No additional significant difference was observed between treatments. Our results indicate that even low concentrations of caffeine, within the range reported in surface waters, may alter zebrafish activity patterns after short-term exposure, with possible ecological consequences for energy allocation and stress-related behaviors. Further studies across concentration gradients are needed to determine dose-response relationships and long-term impacts.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Vulnerability and adaptations of developmental stages to global change stressors

Jeremy DeBonville, Zara-Louise Cowan and Patrice Pottier

Abstract: Anthropogenically induced environmental changes are an increasingly urgent problem, as they are exposing organisms to stimuli and stress beyond what they are adapted to. There is an increasing recognition that certain life stages may be more vulnerable to stressors than others, potentially representing a bottleneck in the vulnerability of species to environmental change. Due to factors including underdeveloped physiological systems and their immobility when faced with challenging environments, embryos have been proposed as being highly susceptible to environmental stressors. Yet, experimental data on this life stage in the context of environmental change is often said to be lacking, in comparison to later life stages. We expect our session to have a broad appeal, to have an interdisciplinary aspect and to bring together speakers and delegates from multiple fields interested in, and working on, the vulnerability and adaptations of this critical life stage to a changing environment. We welcome submissions covering topics relating to vulnerability and adaptations of embryos to environmental stressors (e.g., temperature, acidification, hypoxia, nutrition, toxicants, pathogens), and the impacts that these may have on individuals and populations.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Which fish life stage is most vulnerable to warming?

Cowan, Zara (*zaralouisecowan@gmail.com*) Oral Presentation
University of Gothenburg

Abstract: Climate change is increasing the frequency and intensity of extreme weather events like heatwaves. This is of particular concern for ectotherms such as fish as their basic physiological functions are strongly influenced by environmental temperature. It has been suggested that some life stages of fish are much more vulnerable to temperature changes than others. Specifically, some research has predicted that the upper thermal tolerance limits of embryos and spawning adults are, on average, 10^{o}C lower than larvae and immature adults. If correct, this would mean that climate change is likely to disproportionately impact embryos and spawners, and these life stages should be brought to the top of global conservation priorities. However, these predictions have been criticised for using vastly different measures of thermal tolerance across life stages, potentially underestimating the thermal tolerance of embryos and spawners. Therefore, a critical question remains: which life stages of fish are most vulnerable to warming? To answer this question, we measured the upper thermal limits of four life stages, from embryos to spawners, in eight fish species from around the world using critical thermal methodology.

Results will be discussed in the context of heatwave vulnerability in relation to life history diversity.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Thermal sensitivity of metabolism across the lifespan of a fish

Clark, Timothy (*t.clark@deakin.edu.au*) Oral Presentation
Deakin University

Abstract: The interaction between temperature and metabolism is a key determinant of the responses of ectothermic animals to anthropogenic change. Extreme ends of the lifespan—embryos and spawning adults—are thought to be more sensitive to warming than intermediate life stages due to differential impacts of temperature on aerobic metabolic rates. To test these ideas empirically, we quantify for the first time the thermal sensitivity of metabolism across the lifespan of a vertebrate. Using the black goby (Gobius niger) encompassing a body mass range spanning 5.7 orders of magnitude, we show that the thermal sensitivity of routine metabolism in embryos is low between 8-30oC, contradicting current belief that embryos are especially vulnerable to climate warming due to high metabolic thermal sensitivity. Further, we reveal that aerobic scope, and its thermal dependence, do not meaningfully differ in spawner individuals compared with immature fish, questioning theoretical assumptions that the breadth of aerobic capacity should decline as sexual maturation progresses. We propose that the embryonic life stage of shallow-water fish can be especially robust to acute temperature change, as an adaptation to immobility in the face of environmental variability. Moreover, we suggest that reproductive maturation does not universally impact aerobic scope, and thus any decline in environmental resilience may be driven by other mechanisms. This study shows the importance of experimental research in testing established theoretical assumptions.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Effects of temperature on three life stages of partially armored threespine stickleback

Cooper, Cassidy[1] (*cjcooper@ucdavis.edu*) Oral Presentation
Co-Authors: Zillig, Kenneth [2]; Fangue, Nann [1]
[1] University of California Davis; [2] University of Minnesota

Abstract: Unarmored threespine stickleback (UTS; Gasterosteus aculeatus williamsoni) is an endangered fish native to California (USA) often found in thermally-imperilled streams. We used the closely-related partially armored threespine stickleback (PATS) subspecies as a surrogate species to investigate if certain life stages are more at-risk. We measured CTMAX of adult and juvenile threespine stickleback acclimated to four constant temperatures (15.6, 18.6, 22.7, and 25.0°C, n = 9 - 12 per temperature per life stage). While adults successfully acclimated to all temperatures, juvenile fish showed increased

mortality at temperatures >22.7°C. CTMAX values ranged from 31.3°C - 34.1°C and increased with increasing acclimation temperature for both juveniles and adults. Across all temperatures, adults possessed a significantly higher CTMAX than juveniles (~0.8°C). We also quantified the effects of temperature on larval development and assessed hatching success, time to hatch, and size at hatch for groups of embryos incubated at 15.6, 18.6, 22.7, and 25.0°C. Embryos successfully hatched across all incubation temperatures (n = 37 - 49 viable hatches per temperature). Overall, hatch time decreased as temperature increased, but plateaued at temperatures exceeding 22.7°C. Fish reared at warm temperatures exhibited significantly shorter length at hatch (0.47 cm) than those incubated at 18.6°C (0.49 cm) or 15.6°C (0.52 cm). Understanding differences in thermal requirements across life stages provides critical information for wildlife managers seeking to conserve this species within seasonally variable and thermally challenging environments.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Comparing thermal tolerance of adult and embryonic life stages: Is survival a good measure of tolerance?

Harman, Adam (*harman@zoology.ubc.ca*) Oral Presentation
Co-Authors: Lee, Megan; Schulte, Patricia
The University of British Columbia

Abstract: Accurate assessment of thermal tolerance is essential to understanding and predicting aquatic species capacity to respond to increasing global temperature. Comparisons of acute thermal tolerance between life stages can help determine their relative vulnerability to climate change. However, estimates of tolerance in embryos are generally lacking and usually based on static exposures (e.g. survival) which are largely incompatible with dynamic tolerance estimates (e.g. CTmax). Therefore, it is important to develop more comparable measures of thermal tolerance for embryo and juvenile life stages to better understand the relative susceptibility of life stages to environmental change. Sheepshead minnow (Cyprinodon variegatus) embryos were incubated for the entire duration of embryonic development at 5 temperatures (16, 22, 28, 34, 40°C). Embryos were sampled at Stage 29 and thermal tolerance was quantified with either a modified CTmax protocol, or a static thermal exposure. The CTmax of embryonic sheepshead minnow was similar to that of adults and showed equivalent acclimation capacity. These results will be compared to those derived from static exposures.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Unifying heat tolerance assessments across fish life stages

Pottier, Patrice (*patrice.pottier@bioenv.gu.se*) Oral Presentation
Co-Author: Tomás, Mafalda; Zürcher, Joelle; Jutfelt, Fredrik and Eklund Ella
University of Gothenburg

Abstract: Fish early life stages are widely assumed to be more vulnerable to heat stress than juveniles and adults. However, current comparisons among life stages rely on methods that differ in the choice of endpoints and the duration of heat stress. These methodological inconsistencies have fuelled debates about the higher sensitivity of embryos to heat stress and have limited our ability to predict the impacts of climate change on fish populations. In this study, we compared estimates of heat tolerance obtained from different methods in zebrafish embryos and larvae. We found that integrating the intensity and duration of heat stress results in substantially lower variation in heat tolerance across life stages than previously reported. Our results suggests that the higher reported vulnerability of embryos may not be universal, and may instead be largely explained by methodological inconsistencies among life stages. In addition, we revealed pronounced variation in heat tolerance within embryonic development itself, implying that projections based on a single embryonic time point may under- or over-estimate vulnerability assessments. Together, these results call into question the generality of life-stage differences in heat tolerance, and demand more robust and comparable empirical evidence on fish embryos to assess the vulnerability of fish populations to climate warming.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Temperature-driven embryo vulnerability as a recruitment bottleneck for European smelt (Osmerus eperlanus) in a warming estuary

Harvey, Megan[1] (*megan.harvey@uni-hamburg.de*) Oral Presentation
Co-Author: Dahlke, Flemming[1]; Nikitas Politis, Sebastian[2]
[1]Universität Hamburg, [2]Technical University of Denmark

Abstract: Temperature-driven embryo mortality may represent a critical recruitment bottleneck for fish in temperate estuaries, where steep seasonal thermal gradients are intensifying under climate change. Yet experimental data on this life stage remain scarce, limiting predictions of population responses to warming. Here we show that embryonic mortality sets the lower boundary of the recruitment window in European smelt (Osmerus eperlanus), a declining estuarine fish of conservation concern. We incubated offspring from the Elbe estuary at six temperatures (6–17 °C) from fertilisation to first feeding, measuring hatching timing and success, developmental mortality, routine MO_2 at incubation temperatures and acute increases of +5 and +10 °C, and CTmax using

disorientation and loss of responsiveness as endpoints. We found that hatching onset ranged from day 6 post-fertilisation at 15 °C to day 30 at 6 °C, and the hatching window compressed from 13 to 4 days. As fertilisation occurred at a common temperature, unfertilised egg proportions reflected maternal variation, while post-fertilisation developmental failure increased with incubation temperature. Hatch success peaked at 9–11 °C, reaching 84–87%, and declined to 29% at 17 °C. In contrast, post-hatch survival exceeded 98% across treatments, showing recruitment failure occurs primarily at the embryo stage. In larvae, routine MO_2 plateaued at warm extremes, particularly in older larvae, and non-recovery from the loss-of-responsiveness endpoint reached 48% at 15 °C and 70% at 17 °C. Our results confirm that thermal vulnerability in smelt is stage-specific, with embryos representing a critical life-stage for recruitment and larvae from the warmest treatments facing compounding physiological constraints as their thermal limits are approached.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Early life vulnerability of coregonids to warming: species and population-specific thermal tolerance across Great Lakes and inland lakes

Zillig, Kenneth (*kzillig@umn.edu*) Oral Presentation
Co-Authors: Hansen, Gretchen
University of Minnesota

Abstract: Coregonid fishes are cold-adapted taxa that occupy thermally sensitive habitats across the Northern hemisphere and are vulnerable to climate warming. Early life stages, embryos and larvae, are thought to be especially susceptible to environmental stressors, yet variation in thermal tolerance during these stages remains poorly characterized across species and populations. We collected coregonid embryos from multiple populations and species, including North American Great Lakes populations and an inland lake population from Lake Koronis, Minnesota, and exposed them to incubation temperatures from 4.5 to 16°C to assess differences in embryo thermal tolerance, survival to hatch, and size at hatch. Preliminary results reveal meaningful variation in temperature-dependent embryo mortality and time to hatch between populations and species, suggesting that population-specific tolerance limits likely exist. A consistent theme emerging from this work is that inland lake populations, which exist in more isolated, thermally idiosyncratic environments compared to large lake populations, may harbor distinct physiological traits that are not captured by generalized species-level vulnerability assessments. As global change continues to elevate water temperatures and alter dissolved oxygen regimes, understanding this within-species and within-genus variation will be critical for identifying which populations face the greatest risk and which may possess adaptive capacity. These findings underscore the value of including inland lake populations and species in future assessments of coregonid vulnerability and conservation prioritization.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Can Fish Embryos Acclimate to Warming?

Tomás, Mafalda[1] (*mafalda.tomas@bioenv.gu.se*) Oral Presentation
Co-Authors: Pottier, Patrice[1]; Cowan, Zara-Louise[1]; Gräns, Albin[2] and Jutfelt, Fredrik[1]
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Abstract: Global warming and the rising frequency of aquatic heatwaves are intensifying interest in how ectothermic organisms cope with rapid temperature change. Although thermal acclimation is well documented in juvenile and adult fishes, its scope and limits during embryogenesis remain poorly understood. We investigated how chronic and acute heat exposure during embryogenesis influences thermal plasticity in zebrafish (Danio rerio). Embryos were exposed during developmentto one of six temperature treatments: static temperature (22, 24, 28, 32, 34 °C) or a simulated 36 °C heatwave lasting four hours. Survival to the first feeding stage, hatching success, and hatching time were quantified to define the functional thermal window for development. To evaluate the physiological consequences of early thermal environment, we measured critical thermal maximum (CTmax) and routine metabolic rate.CTmax increased with developmental temperature but plateaued at the highest rearing temperatures, indicating limits to developmental plasticity. In contrast, acute heatwave exposure elicited highly variable responses, with reduced average tolerance in some individuals, suggesting a potential trade-off between short-term hardening and the accumulation of thermal injury. By linking developmental environment to performance and physiology, this study explores whether acclimation provides early life stages with buffering capacity against increasingly frequent thermal extremes.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Physiological effects of chronic elevated CO2 exposure from fertilization to yolk-sac absorption and the freshwater-to-seawater transition in pink salmon (Oncorhynchus gorbuscha).

Lee, Seungyeon (*sylee713@student.ubc.ca*) Oral Presentation
Co-Authors: Bouyoucos, Ian and Brauner, Colin J.
The University of British Columbia

Abstract: Rising global CO2 is reducing environmental pHin seawater (SW) and freshwater (FW) systems. Among Pacific salmonids, pink salmon (Oncorhynchus gorbuscha) leave FW earliest in development (immediately after emergence), making them an ideal model to examine CO2 sensitivity during early development and the FW-to-SW transition.
In this study, we exposed pink salmon immediately following fertilization through to yolk-sac absorption (YSA) to three chronic CO2 exposure treatments: control (400 µatm), medium (2000 µatm), and high (5000 µatm) CO2. To evaluate physiological sensitivity throughout development, we assessed thermal tolerance (critical thermal maximum) and

hypoxia tolerance (incipient lethal oxygen saturation) at the respective CO2 levels at four early developmental stages (600, 711, 808, and 959 accumulated thermal units [ATUs], the latter corresponding with YSA) and at 24 h and 14 d post-SW transfer (990 and 1091 ATUs). Hypoxia tolerance increased during FW development in the medium and high CO2 groups, while elevated CO2 reduced overall tolerance levels. There was no significant change in thermal tolerance during FW development except in the medium CO2 group, where tolerance declined under CO2 exposure. Following SW transfer, CO2-related differences were eliminated in both thermal and hypoxia tolerance.
These results indicate that elevated CO2 exposure in FW significantly reduces physiological tolerance during early development; however, the pattern for the changes differed between hypoxia and thermal tolerance. The diminishing treatment effects after SW transfer indicate that FW stages prior to migration may represent a more CO2-sensitive environment than SW, highlighting stage-specific vulnerability during early development.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Assessing seaweed's potential to buffer ocean acidification in Pacific herring embryos

Chalifoux, Virginie (*chav01@student.ubc.ca*) Oral Presentation
Co-Author: Frommel, Andrea
The University of British Columbia

Abstract: In the coastal waters of British Columbia, CO2 levels can already reach more than 1000 µatm through local oceanographic processes, levels not predicted until the end of the century for the global ocean. These high CO2 levels can impact early life stages of fish, such as Pacific herring. Numerous experiments have demonstrated effects of CO2 leading to larval deformities, delayed development, stunted growth and lower condition, factors that compromise their survival. These experiments have mostly utilized eggs obtained through artificial strip spawning. However, in a recent study, herring embryos collected from the wild attached to seaweed showed no negative effects of CO2 on development, growth and survival. This research aims to evaluate how seaweeds may be able to mitigate negative effects of CO2 on embryonic development through photosynthesis, with carry-over effects on larvae. Recently spawned herring embryos were collected from the intertidal near Squamish, BC on two substrates: naturally occurring seaweeds and cedar, which had been placed by the Átl'ka7tsem/Howe Sound Marine Stewardship Initiative. Embryos were exposed to a CO2 level of 3000 µatm on their respective substrate until hatch and larvae were reared until yolk sac depletion to determine carry-over effects. Embryonic respiration and heart rate were measured, as well as the expression of stress regulation and growth genes. Mortality rate of embryos was 17.6% higher on cedar compared to seaweed when exposed to CO2. This work will provide valuable information for the First Nations who monitor herring spawning and for DFO management for fishing quotas.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

The effect of fluctuating hypoxia on the physiology of larval coho salmon

Osborne, Courtney (*cosb280@mail.ubc.ca*) Oral Presentation
Co-Authors: Chalifoux, Virginie; Belland, Emily and Frommel, Andrea
The University of British Columbia

Abstract: Hypoxia occurs frequently in streams around British Columbia which challenges the developing salmon larvae. Salmon larvae develop in redds, nests up to 300mm deep in the gravel, and significantly fluctuating hypoxia levels are commonly experienced. This study investigates the effect of fluctuating hypoxia on coho salmon (Oncorhynchus kisutch) embryos hatching into larvae. Coho salmon are an important species to study as they are the Pacific salmon species which spends the longest time in rivers and therefore have the potential to be greatly impacted by these conditions. This was done by a month-long exposure to a control 95% dissolved oxygen, 50% dissolved oxygen, or random daily fluctuations to a minimum of 30% dissolved oxygen. The protein content, lipid content, deformities and heart rate were measured weekly. The preliminary results suggest no significant difference between all treatments for mortalities and morphological deformities, however the sub-lethal impacts are still being investigated. This research will be important for understanding the physiological effect embryos may experience in the natural environment and therefore be beneficial for further understanding coho salmon population dynamics.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Impact of Maternally Transferred Methylmercury on Embryo Development and Neurobehavior in Fathead Minnows (Pimephales promelas)

Ortega-Rodriguez, Celeste (*celesteortega-rodriguez@my.unt.edu*) Oral Presentation
Co-Author: Brandon, Emma; Ludbrook, Faith and Roberts, Aaron
University of North Texas

Abstract: Mercury is a non-essential metal of global concern. Emitted by natural and anthropogenic sources into the atmosphere, mercury can be deposited in aquatic ecosystems and be transformed into methylmercury. Methylmercury is a neurotoxic compound and accumulation in aquatic biota from dietary exposure can be associated with adverse health outcomes. In addition, methylmercury can be maternally transferred to offspring and provide an early route of exposure for early life stage (ELS) fish. The maternal transfer of methylmercury can negatively impact development and neurobehavior in ELS fish. Therefore, understanding the embryotoxic effects of maternally transferred methylmercury is crucial for understanding multigenerational methylmercury toxicity. To characterize the impacts of maternally transferred methylmercury, the present study exposed adult fathead minnows (Pimephales promelas) to dietary methylmercury concentrations of 0, 1, and 3 mg/kg methylmercury for 35 days. Adult reproduction

(spawning frequency, clutch size) was monitored daily and embryonic endpoints included mercury accumulation, morphometrics (yolk sac and eye area and embryo size), and heart rate. Preliminary analysis suggests that dietary MeHg exposure may impact spawning frequency in adults but not clutch size. Embryo mercury accumulation increased over time for the two MeHg treatment groups, but no differences were observed between treatments for yolk sac area or heart rate. However, embryonic movement was correlated with MeHg accumulation. The results of the present study will provide insight into the embryotoxicity of maternally transferred methylmercury in ELS fish.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Lasting effects of variation in developmental temperature on later-life hypoxia tolerance

Eisenberg, Rachel (*reisenberg@zoology.ubc.ca*) Oral Presentation
Co-Author: Blanchard, Tessa and Schulte, Patricia
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Abstract: With climate-change, coastal aquatic ecosystems are experiencing dramatic increases in the duration and frequency of warming and hypoxic events, which could have important impacts on sensitive early developmental stages and potentially induce long-lasting phenotypic changes due to developmental plasticity. Here, we explored these questions using Atlantic killifish (Fundulus heteroclitus). Killifish embryos were incubated at 26°C with a diel fluctuation of ± 0, 3, 5, or 7°C from fertilization to hatch, and were then reared at a common, constant, temperature of 18°C for 1.5 years. CTMax was measured at 1-month post hatch and at 1.5 years. Development under the widest fluctuation regime (±7°C) decreased CTMax in 1-month old juveniles, but this effect did not persist later in life. Developmental temperature regime also affected later-life hypoxia tolerance, such that fish that developed at the intermediate and widest thermal fluctuations exhibited time to loss of equilibrium under hypoxia (TLOE; 2% air-sat.) that was ~30% of the TLOE of fish developed under constant temperature, suggesting cross-sensitivity. Differences in TLOE persisted in fish at 1.5 years of age but were only evident when adults were acclimated to 26°C, and not when they were acclimated to 18, or 10°C, demonstrating an interaction between the developmental environment and adult acclimation temperature. The possible role of persistent changes in gene expression underlying these observations will be discussed. These data demonstrate the lasting impacts of altered temperature and thermal fluctuations during early development and potential detrimental effects on interacting phenotypes, highlighting an underappreciated dimension of climate-change impacts on fish.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Early-life stress history determines thermal tolerance in larval zebrafish

Vámos, Sofia[1] (*sofia.duartevamos@ucdconnect.ie*) Oral Presentation
Co-Authors: Wollenberg Valero, Katharina[1]; Jutfelt, Fredrik[2]; Tomás, Mafalda[2] and Braz-Mota, Susana[3]
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Abstract: Extreme heat events are projected to become more frequent, intense, and prolonged over coming decades. Early life stages ectotherms are especially sensitive to such events because their physiology is tightly linked to environmental temperature. Whether early heat exposure increases later vulnerability or instead induces protective plasticity remains unclear. We tested how direct thermal stress and socially mediated chemical cues during early development influence subsequent thermal tolerance in zebrafish (Danio rerio). From 0–5 days post-fertilization, embryos were reared under control conditions, exposed to stress metabolites from heat-stressed donors, or subjected to direct thermal stress. After standard rearing, larvae were either kept at control temperatures or exposed to a second heatwave from 7–10 dpf. Critical thermal maximum (CTmax) was quantified blindly at loss of equilibrium. Thermal tolerance depended strongly on developmental history. A later heatwave increased CTmax in larvae without prior direct heat exposure, including those exposed only to stress metabolites. In contrast, larvae that experienced early direct thermal stress showed reduced CTmax when exposed to a second heatwave, consistent with negative carry-over effects. Mixed-effects models revealed a significant interaction between early environment and later heat exposure, demonstrating that thermal tolerance is shaped by stress history rather than temperature alone. These findings show that embryos are not uniformly vulnerable to warming and that early environmental cues can shape resilience to future heat stress.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Plasticity of cellular and whole animal responses after embryonic thermal and hypoxic stress related to climate change and thermal pollution.

Manzon, Richard G. (*richard.manzon@uregina.ca*) Oral Presentation
University of Regina

Abstract: Climate change and thermal pollution are increasing water temperature, thermal fluctuations, and variability in the fall turnover, ice-cover and spring thaw. We have studied the impact of the embryonic environment on stress responses, thermal tolerance and metabolism of embryos, larvae and juvenile Lake Whitefish (winter spawners), and Yellow Perch and Walleye (spring spawners).Lasting impacts of the embryo environment on aerobic swim performance of whitefish and perch (1-1.5 years) were also studied.Alterations in embryo incubation temperature attenuated the heat shock response

(HSR) to acute stressors in whitefish and perch embryos and post-hatch fish.This plastic response occurred in whitefish at incubation temperatures as low as 3^{0}C above normal and with repeated thermal stress (+3^{0}C for 1h/d).By comparison, it was the median of three environmentally relevant incubation temperatures (12, 15 and 18^{0}C) that attenuated the acute HSR in perch.That both the HSR and aerobic swim performance were altered in juveniles further support the potential for developmental plasticity. Elevated embryonic temperatures reduced critical swim speeds in whitefish tested at and acclimated to15^{0}C but not 21^{0}C.Conversely, routine metabolic rate was lower and aerobic scope higher when tested at 21^{0}C but not 15^{0}C.Aerobic swim performance in perch was not affected.The nature of the cellular stress response to hypoxia in all three species will be discussed.Collective data show that minor alterations in embryonic environments can induce developmental plasticity in temperate freshwater fishes, with these effects being more pronounced in the winter-spawning, stenothermal whitefish than the spring-spawning, eurythermal perch.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

HOW CLOSE IS TOO CLOSE? THERMAL INJURY ACCUMULATION AND RECOVERY NEAR UPPER THERMAL LIMITS IN LARVAE AND ADULT ZEBRAFISH

Braz-Mota, Susana [1] (*susanabmota@gmail.com*) Poster Presentation
Co-Author: Tomás, Mafalda[2]; Lechner, Emily[2], Pottier, Patrice[2]; Vámos, Sofia[3]; Ern, Rasmus[4] and Jutfelt, Fredrik[2]
[1] Instituto Nacional de Pesquisas da Amazônia; [2] University of Gothenburg, [3]University College Dublin, [4]Norwegian University of Science and Technology

Abstract: In ectotherms, growing evidence suggests that upper thermal tolerance is reached when thermal injury accumulate faster than organisms can repair. However, empirical tests of how vertebrates accumulate thermal injury, and how fast they can repair physiological damage remains limited. Here, we tested how the accumulation of thermal injury is impacting the heat tolerance of zebrafish (Danio rerio) larvae and adults, and how well they can repair accumulated injury. Adult zebrafish were acclimated to their critical temperature (37 °C; where injury and repair rates are equal) for seven days and then exposed to 38.5 °C for 24 h (where injury accumulation occurs). Fish were subsequently allowed to recover at 30 °C, where repair is faster than injury, for either 1 h or 5 h before thermal tolerance was assessed. Larvae were exposed to a similar protocol. Thermal tolerance was measured using both fast (0.3 °C/min) and slow (0.03 °C/min) critical thermal maximum (CTmax) protocols. In larvae, exposure to 38.5 °C resulted in clear thermal injury accumulation relative to 37 °C, detected as reduced loss of equilibrium temperature. Interestingly, we detected no recovery within 1 h or 5 h for fast CTmax, but individuals showed signs of recovery after 5h when tested using slower ramping rates. In adults, we found no differences among treatments with fast CTmax, but similar evidence for recovery after 5h for slow CTmax.. Overall, our results show that slow CTmax protocols are more capable of detecting accumulated thermal injury and recovery dynamics,

suggesting different CTmax warming rates may cause loss of equilibrium through partially different mechanisms.Consequently, widely used fast CTmax protocols likely fail the capture the role of injury-repair processes in setting upper thermal limits, and this study continues to add to discussions about the ecological relevance of critical thermal limits.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Thermal performance of early life stage striped bass (Morone saxatilis)

Gaytan, Yvette (*Ygaytan1@students.towson.edu*) Poster Presentation
Co-Authors: Kraskura, Krista
Towson University

Abstract: Thepopulation abundanceof youngstripedbass (Morone saxatilis) has been alarmingly low for the past seven years inthe ChesapeakeBay.Elevatedand variablewater temperaturescancause direct mortalityorseverelyimpactphysiologicalperformance.The rapidly shifting thermal tolerance ofearlydevelopmentalstagesmaydifferdepending onthe durationof temperature exposure. Therefore,ourgoalwastocharacterizethermal limitsof embryonicstriped bass under acute timescalesandlarvalstriped bassfollowingacute and chronictemperatureexposuresbymeasuring theirmetabolic rates(embryos and larvae)andheartrates(larvaeonly).Wemeasured these performances under5temperatures(12ºC, 15ºC, 18ºC, 21ºC,and23ºC).Respirometry wasconductedtoobservethe metabolic rates, and Arrheniusbreakpointtestswere conductedtodeterminecardiac performance.Linking life stages and how thermal performance differs between them will set forth a stronger foundational knowledge of the physiological limitations of early development stages that mayhelpexplain why there has been an abundance decline ofyoung-of-yearstriped bassjuveniles.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Effects of thermal challenge on burbot (Lota lota) egg survival and development

Hubert, Braden (*hubertbraden@gmail.com*) Poster Presentation
Co-Authors: Leonard, Jill and Simmons, Jonathan
Northern Michigan University

Abstract: Burbot (Lota lota) are native to Lake Superior, where they spawn between January and March in shallow areas or tributary rivers when water temperature is 1°C to 4°C. Burbot are broadcast spawners with females releasing between 45,000 and 3 million eggs per season, with hatching 30 to 100 days post-fertilization. Studying the effect of temperature on development is important for understanding burbot resilience to warming water conditions. This experiment investigates the effects of 3°C, 5°C, 10°C, and 15°C exposures on egg development and survival in a sample of burbot spawned from the Chocolay River in Marquette, Michigan, in January 2026. Fertilized eggs were transferred to the laboratory for

maintenance at 3°C, and random groups of 10+ eggs were moved to 3°C, 5°C, 10°C, and 15°C every 48 hours throughout development. For each trial, the number of surviving eggs was recorded every 24 hours for five days after transfer. Overall, the 10°C and 15°C trials had significantly higher mortality rates than the 3°C and 5°C trials, which suggests a negative effect of warmer temperatures on egg survival and development. Egg resiliency to warmer temperatures increased throughout development, with survival percentage increasing over time. Additionally, the developmental stages of segmentation and near-hatching showed substantial increases in resilience to warmer temperatures when compared to pre-segmentation. This confirms prior work showing that low initial rearing temperatures are necessary for embryonic development and suggests that planktonic drift into warmer areas may be deleterious during the embryonic stages for this species.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Intermittent Hypoxia Modulates Morphological and Cardiorespiratory Phenotypes in Developing Zebrafish (Danio rerio)

Horak, Bailey[1] - Poster Presentation
Co-Authors: Martinez Bautista, Gil[1]; McAden, Laney [2]; Ho, Annabelle[1]; Shah, Rian[1] and Burggren, Warren[1]
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Abstract: In natural freshwater environments dissolved oxygen often fluctuates, making intermittent hypoxia 'more' ecologically relevant than constant hypoxia for many freshwater species. Here, developing larval AB wild-type Zebrafish (Danio rerio) were used to assess how intermittent and continuous hypoxia modulate growth rate, cardiac function (heart rate, stroke volume, output), and hypoxia tolerance as time to loss of equilibrium (TLoE). Fish were exposed from fertilization to 7 days post-fertilization (dpf) to normoxia (controls; PO2 ~21 kPa), intermittent hypoxia (12h normoxia/12h hypoxia ~10 kPa), or continuous hypoxia (PO2 ~10 kPa). Generally, body mass was not significantly different among groups during development to 7 dpf, but yolkless embryo mass was consistently higher in control fish. Specific growth rate in control larvae was 10-17% daily compared to 6-14% in intermittent hypoxia fish, and 8-14% in continuous hypoxia fish. Heart rate was ~151, ~141, and ~129 beats min-1 in control, intermittent, and continuous hypoxia larvae, respectively, while stroke volume was ~0.29, ~0.20, ~0.16 nL beat-1, respectively. Calculated cardiac output was ~44, ~29 and ~22 nL min-1, respectively. Hypoxia tolerance measured as TLoE was ~2.5 min in control fish but higher in intermittent and continuous hypoxia groups (~5.3 and ~6.0 min, respectively). Collectively, our data indicates that intermittent and continuous hypoxia decrease cardiac performance and delay early growth in zebrafish, yet they concurrently enhance larval hypoxia tolerance. This suggests a trade-off where early-life oxygen availability promotes survival under low oxygen at the cost of cardiovascular capacity and growth.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Effects of developmental stage on thermal tolerance and thermal preference of larval burbot (Lota lota) and lake whitefish (Coregonus clupeaformis)

Simmons, Jonathan (*jonsimmo@nmu.edu*) Poster Presentation
Co-Author: Leonard, Jill
Northern Michigan University

Abstract: Anthropogenic climate change poses a particular risk for cold stenothermal species such as burbot (Lota lota) and lake whitefish (Coregonus clupeaformis) that hatch in early spring and require near-freezing water during embryonic and early larval development. Climate warming could impact these species' geographic ranges or affect embryonic and larval survival in habitats with different thermal properties; previous research with burbot and lake whitefish has shown that higher incubation temperatures are correlated with greater mortality and a decrease in time-to-hatch. However, while prior work with these species or closely related species has indicated a capacity for thermal plasticity, limited information is available about how the thermal tolerance, thermal preference or acclimation capacity of burbot or lake whitefish vary across different larval developmental stages. This project uses critical thermal maximum (CTmax) testing and thermal preference maze testing of larval burbot and lake whitefish across multiple points in the yolk sac, preflexion and flexion stages to investigate stage-dependent differences in the thermal tolerance and preference of these two cold-water benthic species, both of which play significant roles in the ecology of the Great Lakes region.

DEVELOPMENT Vulnerability and adaptations of developmental stages to global change stressors

Molecular Signatures of Early Stress Responses to Emerging Contaminants

Bertucci, Juan Ignacio (*jignacio.bertucci@ieo.csic.es*) Poster Presentation
Co-Author: Martínez Schönemann, Alexandre and Bellas Bereijo, Juan
Centro Oceanográfico de Vigo

Abstract: Emerging contaminants such as citalopram and triclosan are increasingly present in coastal ecosystems, where they may compromise biological functions essential for organismal performance and ecosystem stability. Their effects gain relevance when combined with projected climate-driven conditions such as ocean warming and ocean acidification, which can modify physiological responses and alter the sensitivity of early developmental stages. Understanding how these drivers jointly modulate phenotypic and molecular traits is key for improving mechanistic predictions of organismal tolerance under future environmental scenarios. In this study, larvae of Paracentrotus lividus were exposed for 48 h to environmentally realistic concentrations of citalopram and triclosan under simulated warming and acidification. Phenotypic descriptors and transcriptomic profiles were quantified to identify stress-responsive biological processes and evaluate whether

molecular alterations anticipate functional impairments. Growth remained stable under most climate treatments, yet both contaminants induced marked transcriptional reprogramming even when morphological endpoints were unaffected. Citalopram mainly altered pathways involved in metabolic regulation, early development, and cellular defence, whereas triclosan affected processes related to microtubule organization, energy metabolism, and calcium-dependent signalling. Combined exposures often intensified these responses, revealing that multiple drivers can generate biological outcomes that differ considerably from those produced by individual stressors. These findings underscore the importance of integrating high-resolution molecular tools into functional biology, as they can detect early disruptions that may not be evident through conventional phenotypic assessments. By linking developmental performance with stress-responsive pathways, this work contributes to a more mechanistic understanding of how coastal species may sustain, or lose, functional resilience under future environmental pressures.

IMMUNOLOGY Fish immunology in a changing environment: Implications for aquaculture and conservation

Fish immunology in a changing environment: Implications for aquaculture and conservation

Mark Fast and Ensiyeh (Forough) Ghanizadeh-Kazrouni

Abstract: Fish are increasingly exposed to a wide range of infectious and non-infectious stressors including pathogens, pollutants, rising temperatures, hypoxia, and habitat degradation. These stressors can have profound impact on immune function, resulting in increased disease susceptibility, compromised fitness, and long-term health consequences. Given that food security and climate change are two of the most pressing challenges globally, this session will focus on how environmental changes shape immune responses and adaptation mechanisms in ecologically and economically important species. The topics of interest will cover role of stressors in inducing immune response, emerging biomarkers of immune stress, and the application of transcriptomic and proteomic technologies for monitoring fish health and remediation strategies. This session aims to bring together researchers from immunology, evolutionary biology, and physiology to meet and share discoveries and ideas, and foster collaborations.

IMMUNOLOGY Fish immunology in a changing environment: Implications for aquaculture and conservation

Spatial transcriptomic analysis of gill regeneration in Atlantic salmon using Hi-Plex RNAscope

Ghanizadeh-Kazerouni, Ensiyeh[1] (*Forough.ghanizadeh@ubc.ca*) Oral Presentation
Co-Authors: Frasca Jr., Salvatore[2]; Fast, Mark D.[3] and Brauner, Colin J.[1]
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Abstract: Atlantic salmon gills exhibit remarkable regeneration capacity, which necessities coordinated responses. Although transcriptomic studies have identified genes associated with gill regeneration, little is known about the spatial distribution of these markers within distinct cellular compartments. This study used Hi-Plex RNAscope to characterize spatial expression patterns of key genes associated with gill regeneration. Fish were subjected to 50% resection of a few filaments on middle and ventral regions of first gill arch. The mRNA expression intensity for mmp-14, il-1β, mucin-5a, and notch (involved in epithelial proliferation, immune activation, and signalling pathways) was quantified in the tip of resected/regenerating and internal-control filaments (ICFs), at 1, 4, and 12 week-post-resection (wpr). Il-1ß and mucin-5a transcripts were detected in ~13% of cells, whereas notch and mmp-14 were expressed in 48-54% of cells. Il-1ß transcript abundance was

significantly higher in resected/regenerating filaments across all time points, indicating a sustained pro-inflammatory response to injury. Compared with ICFs, the mRNA abundance of mmp-14 in resected/regenerating filaments was higher at 1 wpr, consistent with early extracellular matrix remodeling and cell migration, whereas mucin-5a was elevated at 4 wpr, suggesting a delayed epithelial response. While il-1ß and mmp-14 did not display cell-specific expression, mucin-5a was predominantly expressed in mucus cells. In contrast, notch mRNA levels were higher in ICFs than resected/regenerating filaments and showed a clustered expression pattern that appeared to be associated with ionocytes, independent of resection. This suggests that notch may function as a lineage-specific signaling pathway and a pre-existing mechanism to regulate tissue homeostasis and regeneration.

IMMUNOLOGY Fish immunology in a changing environment: Implications for aquaculture and conservation

Salmonid red blood cells act as sensors of stress and infection

Dahle, Maria (*maria.dahle@vetinst.no*) Oral Presentation
Co-Author: Tsoulia, Thomais
Norwegian Veterinary Institute

Abstract: The red blood cells (RBCs) of Atlantic salmon are multifunctional nucleated cells that can respond to extracellular signals by altering gene expression and function. Genes expressed in resting salmonid red blood cells indicate a multitude of putative functions in addition to oxygen transport. Including: Genes encoding pathogen-sensing pattern recognition receptors, associated signaling mediators, transcription factors and antiviral effector genes, cytokine and chemokine receptors, antigen processing and presentation pathways, as well as receptors for stress hormones.
The dsRNA orthoreovirus piscis (PRV) infects and replicates in RBCs in aquacuture salmon prior to infection of heart and red skeletal muscle myocytes and onset of heart and skeletal muscle inflammation (HSMI). PRV and poly (I:C) dsRNA strongly trigger expression of antiviral response genes in isolated RBC. In vivo, a delay and differential expression profile in the response to PRV infection can be associated with PRV genotypes that trigger a more pathogenic outcome. This indicates a functional readout from red blood cells that may be linked to salmon resilience to PRV infection.
Red blood cells express glucocorticoid receptor (GR) genes at high levels and respond strongly to GR agonists and cortisol ex vivo and in vivo by inducing sensitive target genes. Activating GR in RBCs has a notifiable inhibitory effect on their antiviral responses and genes involved in antigen processing pathways. Certain sensitive GR target genes in RBCs have a promising potential as novel stress biomarkers in salmon blood.
MinIon-Oxford Nanopore sequencing technology have been adapted for monitoring of promising RBC biomarkers in aquaculture salmon.

IMMUNOLOGY Fish immunology in a changing environment: Implications for aquaculture and conservation

Cut it out! Functional genomics of sea lice resistance in CRISPR-edited Coho salmon

Lo, Javon[1] (*jlo9398@upei.ca*) Oral Presentation
Co-Authors: Semple, Shawna L.[1]; Ostbye, Tone-Kari[2]; Fast, Mark D.[1]; Krasnov, Aleksei[2]; Tengs, Torstein[2]; Tilahun Bizuayehyu, Teshome[2] and Robinson, Nicholas[2]
[1]University of Prince Edward Island, [2]Nofima

Abstract: Sea lice parasitism remains the biggest pathogenic threat to salmonid farming in the Northern hemisphere. Sea lice (Lepeophtheirus salmonis) cause open lesions to the skin and fins of salmonids. Chronic infestations can impact growth, homeostasis, and eventually lead to death. Salmonid species display varying susceptibility to sea lice. Coho salmon (Oncorhyncus kisutch) are naturally resistant and capable of rapidly eliminating juvenile parasite stages, while Atlantic salmon (Salmo salar) remain highly susceptible. Previous transcriptomic studies identified immune genes highly expressed in lice-infested Coho salmon but are comparatively unaltered in infected Atlantic salmon. We used CRISPR/Cas9 gene-editing to generate novel Coho salmon knockouts (KO) targeting those candidate genes to functionally annotate their importance in lice resistance. This work is the first successful generation of gene KO in Coho salmon. Edited Coho embryos were grown to smolt stage (~1 year post fertilization) and subject to an infestation trial with L. salmonis copepodids, where gene-edited Coho salmon demonstrated increased lice settlement and development on the fin compared to wild type. Functional KO were validated by Next-Seq and qPCR, confirming that gene disruption impaired sea lice rejection. Target-specific KO differentially altered leukocyte trafficking within 24 hours of exposure, confirming their role in localization and louse rejection, while potentially impacting other immune processes. Lastly, RNA-seq quantification of the fin transcriptome is expected to provide a more complete picture of the altered Coho salmon immune response. The findings from this work will establish the foundational framework for introducing innate sea lice resistance into the otherwise susceptible Atlantic salmon.

IMMUNOLOGY Fish immunology in a changing environment: Implications for aquaculture and conservation

Effects of temperature on host immunity and disease severity in salmonid bacterial kidney disease

Wheatley, Taylor (*twheatley@upei.ca*) Oral Presentation
Co-Author: Semple, Shawna L.; Whyte, Shona K. and Fast, Mark D.
University of Prince Edward Island

Abstract: The unpredictable hydrodynamics of aquatic systems have profound impacts on aquaculture operations, creating stress for species confined to net pens. These environmental stressors can hinder production by reducing feed consumption, impacting growth and harvest timing, but more notably, increasing susceptibility to infectious disease. While vaccination and antibiotics represent typical mitigation strategies for

farmed fish, these interventions are expensive, can contribute to antimicrobial resistance, and have limited effectiveness or sustained protection. Recent studies identified that behavioural thermoregulation may enhance host survival by regulating inflammatory responses and pathogen clearance during infection. These findings could help target pathogen-specific responses, which can be exploited within controlled aquaculture settings to improve the management of disease outbreaks. In particular, Renibacterium salmoninarum, an intracellular bacterium responsible for the chronic infection in salmonids known as bacterial kidney disease, is of growing concern as this pathogen continues to appear on fish farms worldwide. This work aims to explore temperature manipulation as a natural, more sustainable approach to improve immune response in commercially important farmed species. To examine these effects, Atlantic salmon acclimated to either 13°C or 18°C were injected intraperitoneally with R. salmoninarum. Throughout the study, blood and tissue samples were collected to assess antibody production, gene expression and infection level. Encouraging results show that the fish responded positively to the thermal treatment, as evidenced by improved survival and reduced bacterial load at 18°C compared to 13°C. These findings provide valuable insights into the temperature-dependent effects on immune response and show a promising, more sustainable future for the global aquaculture industry.

IMMUNOLOGY Fish immunology in a changing environment: Implications for aquaculture and conservation

Integrating Host Immunity and Environmental Surveillance to Manage PKD in Rainbow Trout Aquaculture

King Wolf, Lisa[1] (*Lisakingwolf@gmail.com*) Oral Presentation
Co-Authors: Hartikainen, Hanna[1]; Holland, Jason[2]; Robinson, Oliver[3]; and Hewage, Tharindu[2]
[1]University of Nottingham, [2]University of Aberdeen, [3]British Trout Association

Abstract: The future of sustainable aquaculture is continuously challenged by emerging infectious diseases and climate change. One of these challenges is Proliferative Kidney Disease (PKD), caused by themyxozoanparasiteTetracapsuloidesbryosalmonae. Development of PKD is highly temperature sensitive andthe diseasehasemergedas a significant parasitic diseaseaffecting rainbow trout aquacultureacross Europe and USA. Pathogeninfection pressure was profiledovertwoyearsusingenvironmental DNA (eDNA)monitoring(qPCR)in the inlets offour UK farms.Infection pressureiscorrelated with river-water temperature, waterflowand primary host prevalencetomodelpathogen seasonality.Immune profile development in fishrelativeto infectionisevaluated using a parasite antigen-specific ELISA infish exposed at different timepoints.
Parasite-specific antibody responses beginto develop at 12-14 weeks post natural parasite exposure, with peak production occurring after 17-19 weeks. Notably, antibody levels persist through the winter and decline during earlyspring, beforere-exposureto the parasiteas their prevalence increases.eDNA-derived parasite and primary parasite host concentrations showed clear seasonality, with antibody responses following

pathogenseasonalitypressure.These patternsdemonstratethat the timing of exposure is crucial for developing natural immunity and resistance to PKD during the following summer.
Coupling the eDNA detection(infective dose)with the immunological profiling (fish response to infection)providesadata-driven method to optimise managementstrategiessuch aspre-exposure challenge programs.Integration of seasonality and inter-annual variation in weather with PKD dynamics enablesthedevelopment of predictive models thattransformadaptivemanagement strategies and aquaculture sustainabilityin inland trout aquaculture.

IMMUNOLOGY Fish immunology in a changing environment: Implications for aquaculture and conservation

The effect of chronic CO2 exposure on the growth, metabolic rate and immune response of juvenile Lake Whitefish (Coregonus clupeaformis)

Fajei, Eyesun[1] (*eyesun.fajei@utoronto.ca*) Oral Presentation
Co-Author: Porteus, Cosima [1]; Huyben, David [2]; Hoogenboom, Lisa [2]
[1] University of Toronto Scarborough; [2] University of Guelph; [3] Bamfield Marine Sciences

Abstract: Elevated CO_2 and aquatic acidification, particularly in intensive aquaculture, may threaten the survival and production of Lake whitefish (Coregonus clupeaformis), a species of ecological and economic importance in Ontario's Great Lakes. This study investigated the effects of elevated CO_2 on stress, growth, immunity, and bioenergetics in lake whitefish reared under controlled recirculating conditions at OARC. Fish (n = 249; 71.71 ± 1.26 g) were exposed to four CO_2 concentrations (5, 10, 15, and 20 mg/L) for 16 weeks following a two-week acclimation while fed a commercial diet (3 mm Blue Water). Growth, somatic indices (HSI, VSI), body composition, and metabolic traits (SMR, MMR, AAS) were measured. Immune responses were assessed 24 h after poly(I:C) or saline injection, followed by head-kidney gene expression analysis. Elevated CO_2 significantly reduced thermal growth coefficient (P= 0.02, P = 0.001; high vs. control and low), specific growth rate (P = 0.01, P = 0.0002), and weight gain (P = 0.004, P = 0.00008). Final mass, fork length, somatic indices, and metabolic rates did not differ among treatments. Feed conversion ratio increased under high CO_2 compared to low (P = 0.03), while proximate composition (protein, moisture, ash) remained stable. Compared with commonly cultured species such as Rainbow trout and Nile tilapia, which tolerate higher CO_2, lake whitefish appear more sensitive to CO_2 elevation. These findings highlight the challenges of aquatic acidification in intensive culture and support targeted management and nutritional strategies to maintain growth, health, and production efficiency in lake whitefish aquaculture.

IMMUNOLOGY Fish immunology in a changing environment: Implications for aquaculture and conservation

“Effect of florfenicol and oxytetracycline on the expression of proinflammatory genes in SHK-1 cell line from Salmo salar: focus on histone acetylation"

Morera, Francisco J.[1] (*fmorera@uc.cl*) Oral Presentation
Co-Authors: Rovegno, María Ignacia [1]; Vargas-Chacoff, Luis [2]; Zambrano, Angara[2]; Chipon, Carina[2]; Nualart Carrasco, Daniela Paz[2]; Muñoz, Jose Luis[3]
[1] Pontificia Universidad Catolica de Chile; [2] Universidad Austral de Chile, [3]Universidad de los Lagos

Abstract: Salmon farming conditions often lead to an increased incidence of various bacterial infectious diseases, necessitating the use of antibiotics to maintain fish health. However, the excessive use of antibiotics has become a significant issue within this industry in Chile.
Our research group has recently shown that different concentrations of oxytetracycline and florfenicol can alter the expression of immune-related genes in the SHK-1 cell line, derived from the anterior kidney of Atlantic salmon. Despite these findings, the effects of these antibiotics on epigenetic marks, such as histone modifications, in Atlantic salmon remain unknown.
In this study, we aimed to investigate whether the effects of these antibiotics on the gene expression of innate immunity biomarker genes (TLR-1, MyD88, and INFγ) are associated with changes in histone modifications, as a molecular mechanism linked to genetic changes. We studied the epigenetic effects of florfenicol and oxytetracycline at the cellular level, focusing on changes in histone acetylation.
Using the SHK-1 cell line as a model, we analyzed immune biomarker expression by quantitative polymerase chain reaction (qPCR). We assessed global histone acetylation by Western blot. Our findings indicate that antibiotic treatment induces not only significant changes in the expression of innate immunity biomarker genes, but also alters histone H3 lysine 9 acetylation (H3K9ac) levels. These results suggest that antibiotic treatment can alter the immune response, potentially through epigenetic mechanisms.
This work was supported by Fondecyt 1250678 (LVCh), ANID-Millennium Science Initiative Program-Center ICN2021_002.

EVOLUTION Fish evolution in human-altered environments

Fish evolution in human-altered environments

Andrew Whitehead

Abstract: Human induced environmental changes, such as pollution, harvest pressure, invasive species, and climate change, are posing challenges to the persistence of wild fish species worldwide. Many environmental changes are sufficiently severe to exceed physiological, behavioral, or demographic compensatory abilities, such that fitness is threatened. This symposium will examine the causes and consequences of evolutionary change in the Anthropocene, including research on the features of fish species or the environment that enable or disable adaptive outcomes, the genetic and physiological mechanisms of rapid evolutionary change, and the costs and limits of adaptation.

EVOLUTION Fish evolution in human-altered environments

Evolutionary rescue from climate warming - insights from two artificial selection experiments

Jutfelt, Fredrik[1] (*fredrik.jutfelt@bioenv.gu.se*) Oral Presentation
Co-Authors: Silva-Garay, Lorena[2]; Morgan, Rachael[3]; Pélabon, Christophe[2]; Leeuwis, Robine[2]
[1]University of Gothenburg, [2]Norwegian University of Science and Technology, [3]University of Bergen

Abstract: Attempts to quantify evolutionary potential have been made using various breeding designs, yet artificial selection experiments remain the gold standard. We conducted two artificial selection experiments in populations of wild-caught zebrafish.
In the first experiment, we selected for seven generations to increase or decrease acute upper thermal tolerance, with an additional group selected for increased plasticity in thermal tolerance. We found that thermal tolerance increased by 0.04°C per generation, which is likely insufficient to prevent impacts from warming in most fish species. In fish selected for increased plasticity we instead detected a decrease in plasticity, a sign of a trade-off between adaptation and acclimation.
In our second experiment, we attempted to diverge thermal performance curves (TPCs) to shift up and down in temperature. Over six generations, we selected for high growth rates at low, optimal, or high temperature, which was hypothesized to select for the temperature-specific components of growth. Indeed, the growth rates increased in all lines at their selected temperatures, and were relatively lower at the non-selected temperatures. This meant that the TPCs had evolved in the predicted directions. We additionally traced the underlying physiological mechanisms to increased feed conversion efficiency and reduced energy expenditure at the temperatures fish were selected to perform well at.

We argue that selection experiments are crucial for understanding the potential for, and the mechanisms behind, evolutionary rescue to environmental stressors.

EVOLUTION Fish evolution in human-altered environments

Slow science, fast evolution: Thermal performance curves evolve through artificial selection in zebrafish

Leeuwis, Robine H.J. (*robin.leeuwis@ntnu.no*) Oral Presentation
Norwegian University of Science and Technology

Abstract: Adaptive evolution may allow ectotherms to cope with the negative impacts of climate change through shifts in thermal performance curves, but whether such responses are rapid or slow remains unclear. Artificial selection experiments are a powerful tool for studying adaptation, yet are rarely applied to vertebrates due to their long generation times. Here, we used zebrafish to investigate whether thermal performance curves for growth are evolvable in a vertebrate ectotherm. Over six generations, we selected fast-growing juveniles reared at cold, optimal, or warm temperatures – without directly selecting on the thermal performance curve itself. We phenotyped >12,000 fish over >2 years, unprecedented for a vertebrate selection experiment, and assessed whether changes in feed efficiency and energy expenditure contributed to any evolutionary response.
Our data show a strong realised response to selection across all treatment temperatures, with growth rates improving by ~0.07%/day per generation, suggesting thermal growth performance is highly evolvable. Cold and warm exposure initially reduced growth rates from 1.2 to 0.8%/day relative to optimal, but by F5 the original growth rate had been fully recovered. Thermal performance curves left- and right-shifted in cold- and warm-selected lines, with warm-selected fish outgrowing cold-selected fish at warm temperatures by F3. Better feed conversion efficiency and lower swimming energy expenditure were linked to faster growth under both conditions.
These results demonstrate that thermal performance curves can rapidly evolve as an indirect response to growth selection, advancing our understanding of vertebrate adaptation to climate change and underscoring the role of selection in shaping thermal resilience.

EVOLUTION Fish evolution in human-altered environments

The ecology of gene expression in response to temperature in threespine stickleback

Rogers, Sean[1] *srogers@ucalgary.ca* Oral Presentation
Co-Author: Stanford, Brenna C.M. [2]
[1] University of Calgary; [2] Wilder Institute

Abstract: Gene expression ultimately links DNA to function, yet ecologists have only recently begun to test evolutionary predictions associated with this molecular phenotype. Temperature is a predominant abiotic factor shaping biodiversity through effects on physiology and phenotypic plasticity. Early-life thermal stress may have immediate effects

on performance or induce persistent developmental plasticity expressed later in life. We tested this hypothesis in threespine stickleback (Gasterosteus aculeatus), predicting that early thermal environments generate genotype-by-environment responses mediated by gene expression. Experimental crosses were split among three developmental temperature treatments—acute low (12 °C), control (15.5 °C), and high (21 °C)—and reared to hatch before all individuals were raised to adulthood at a common temperature. We measured temperature-associated phenotypes and conducted 3' mRNA sequencing in muscle and liver. Early-life temperature stress induced consistent changes in cold but not heat tolerance, with lasting effects on gene networks indicating that developmental temperature irreversibly altered aspects of thermal performance

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EVOLUTION Fish evolution in human-altered environments

The presence of domesticated alleles reduces survival and influences the phenotypes of rainbow trout in a semi-natural environment.

Schulte, Patricia (*pschulte@zoology.ubc.ca*) Oral Presentation
Co-Author: Earhart, Madison
The University of British Columbia

Abstract: Domestication is often associated with changes in the genotypes and phenotypes that can reduce the fitness of domesticated animals in natural environments. To examine the effects of domestication on fitness-related phenotypes and genotypes in rainbow trout (Oncorhynchus mykiss), we generated F2 hybrids between a domesticated and a wild strain, and stocked them into two replicate naturalized ponds shortly after hatching. After rearing in the ponds for 6 months, juveniles were trapped and hypoxia tolerance, weight, length, and condition factor were assessed, and tissue samples were collected. Of the originally stocked 10,000 fish, only 302 fish were trapped across both ponds, and 29 additional individuals were captured after the application of the pesticide rotenone, indicating intense selection in the ponds. Using a 200,000 single nucleotide polymorphism (SNP) chip derived from whole-genome sequences of wild and domesticated trout populations from across the species range, we identified alleles characteristic of the domesticated strain and investigated the influence of these alleles on phenotypes and pond survival. There was evidence of selection against domesticated genotypes, as fish with wild genotypes had higher survival in the ponds than those with domesticated alleles. Further, the proportion of domesticated alleles impacted various phenotypes including condition factor. These data highlight the potential deleterious effects of domestication on the fitness of rainbow trout in the wild and provide insight into the genetic basis of this effect.

EVOLUTION Fish evolution in human-altered environments

Evolved transcriptomic responses to multiple pollutant classes in Atlantic killifish

Rios, Carolina[1] (*crios@ucdavis.edu*) Oral Presentation
Co-Authors: Karchner, Sibel[2]; Aluru, Neel[2]; Burke, Tara[3]; Roach, Jen[1]; De La Torre, Ashley[4]; Hahn, Mark[2]; Clark, Bryan[3] and Whitehead, Andrew[1]
[1] University of California Davis; [2] Woods Hole Oceanographic Institution, [3] Canadian Environmental Protection Agency, [4] Oregon State University

Abstract: The composition of pollutants in urban estuaries varies based on historical and contemporary industrial activity and imposes strong selection pressures on urban estuarine organisms. In these environments, populations may evolve resistance to multiple contaminants, particularly when contaminants share a mechanism of action. Understanding the molecular basis and specificity of resistance is critical for predicting evolutionary outcomes in response to environmental change. Atlantic killifish, or mummichog (Fundulus heteroclitus) populations inhabit highly polluted urban estuaries and have rapidly and repeatedly evolved resistance to exposure-induced developmental cardiotoxicity. Atlantic killifish have evolved resistance in both dioxin-like compound (DLC)-dominated and polycyclic aromatic hydrocarbon (PAH)-dominated urban environments. These two chemical classes exhibit both shared and unique mechanisms of toxicity. To investigate the complexity of the molecular mechanisms underlying pollution resistance, we examined transcriptomic responses to two cardioteratogenic pollutants during embryonic development: 3,3',4,4',5-pentachlorobiphenyl (PCB-126, a DLC) and benzo[k]fluoranthene (B[k]F, a PAH). We compared transcriptomic responses between four populations, including two from heavily contaminated sites (PCB-contaminated and PAH-contaminated) with evolved resistance and two from nearby reference sites. For both pollutants, F2 embryos were assessed for a set of developmental heart defect phenotypes across a concentration range, coupled with RNA-seq analysis. Resistant populations demonstrated reduced cardiotoxic phenotypes in response to both classes of pollutant exposure relative to sensitive populations. Comparative transcriptomics revealed shared molecular mechanisms underlying pollution resistance dependent on chemical class (PCB or PAH) and genetic background. This work highlights how complex exposure histories can result in conserved evolved molecular responses and phenotypic outcomes in a wild species.

EVOLUTION Fish evolution in human-altered environments

Evidence of evolutionary adaptation following a series of environmental and demographic disasters in Pacific herring

Whitehead, Andrew[1] (*awhitehead@ucdavis.edu*) Oral Presentation
Co-Author: Tisthammer, Kaho [2]
[1] University of California; [2] University of Hawaii

Abstract: The causes and consequences of population collapse and recovery are often difficult to identify and predict because environmental drivers may interact in complex

ways. The Prince William Sound (PWS) stock of Pacific herring famously collapsed in the early 1990s following the Exxon Valdez oil spill (EVOS), yet both the causes of the collapse and reasons for a persistent lack of recovery remain controversial. Three spawning seasons after the spill, the PWS population declined precipitously, coincident with repeated disease outbreaks, and has remained depressed for more than three decades. We compared relative sensitivity to oil exposure and virus exposure among populations. The PWS stock was not more sensitive than others to these exposures. As a forensic tool, we tracked genome-wide genetic variation through time, spanning the period immediately after the spill but preceding the collapse and extending across three subsequent decades, in the PWS population and two reference populations from Alaska. We found no evidence for loss of genome-wide genetic diversity. However, sensitive temporal analyses revealed significant signatures of polygenic adaptation in the PWS population but not in either reference populations. Identity of candidate genes were consistent with selection acting on immune function and pathways that regulate crude oil developmental toxicity in the period following the spill and collapse. Together, these results suggest that environmental perturbations unique to PWS, including oil exposure and disease outbreaks, affected the fitness of resident herring and shifted their evolutionary and demographic trajectory.

EVOLUTION Fish evolution in human-altered environments

Evolutionary relationships among lake whitefish in the upper Laurentian Great Lakes

Gillespie, Jonah (*jongille@nmu.edu*) Poster Presentation
Co-Author: Diefenbach, Michael and Mandeville, Elizabeth
Northern Michigan University

Abstract: Identifying evolutionary relationships among populations and sympatric species is a foundational goal of systematic biology and is critical for conserving biodiversity in changing environments. Coregonines (whitefishes and ciscoes) in the upper Laurentian Great Lakes represent a recent post-glacial radiation that has experienced severe population declines, extirpations, and ecological disruption due to climate change, invasive species, and fishing harvest pressure. However, evolutionary relationships and demographic histories (population size changes through time) of many populations—particularly inland-lake cisco forms and Lake Superior lake whitefish—remain unresolved. We used population genomic data and demographic modeling to characterize evolutionary relationships, historical admixture, and recent demographic histories of lake whitefish (Coregonus clupeaformis) from four populations in the upper Laurentian Great Lakes. Reduced-representation genomic sequencing (genotyping by sequencing, GBS) were used in phylogenomic, admixture, and demographic analyses to resolve evolutionary lineages, construct phylogenies, estimate divergence timing, and reconstruct population changes in relation to recent anthropogenic disturbances. Results clarify taxonomic uncertainty, identify evolutionarily significant metapopulations, and provide historical context for conservation and management agencies, while broadly contributing to the literature on vertebrate evolution. Future analyses for this project will include data from more Lake

Superior populations of lake whitefish, as well as populations of inland lake cisco ecotypes.

EVOLUTION Fish evolution in human-altered environments

The Osmorespiratory Compromise in Freshwater Prickly Sculpins (Cottus asper)

Liu, Bing (*lbz0802@student.ubc.ca*) Poster Presentation
Co-Author: Liu, Shuang and Richards, Jeffrey
The University of British Columbia

Abstract: Fish are thought to have arisen in seawater (SW) and subsequently invaded freshwater (FW). With the expansion of their ecological niches, the transition to FW necessitates changes in ion regulation to minimize ion loss. Previous data suggested that FW adaptation in prickly sculpins (Cottus asper) is associated with a smaller gill, which may help preserve internal ions in FW compared to their SW sister species. The smaller gill morphology seen in FW, however, may result in a functional conflict between ion regulation and gas exchange (i.e. an osmorespiratory compromise). The goal of this work was to determine whether FW adaptation may be associated with an osmorespiratory compromise.I used populations of prickly sculpins from three regions that differ in FW residence time (from shortest to longest: coastal rivers CRV, coastal lakes CLK, and interior lakes ILK), acclimated them to SW, FW, and ion-poor water (IPW), and assessed gill morphology and respirometric variables. Total gill surface area of fish from the different regions responded differently to acclimation conditions, such that SW exposure increased gill surface area relative to FW and IPW in the CRV populations and had no effect in the CLK and ILK populations.The CRV population had a higher maximum metabolic rate (MMR) and a larger aerobic scope (AS) compared with the ILK population. The CRV population was more hypoxia-tolerant than the ILK population.Overall, these data suggest that longer FW residence time in the ILK population is associated with reduced aerobic capacity, consistent with greater osmorespiratory compromise.

IONS I Ion and acid-base regulation in fishes

Ion and acid-base regulation in fishes

Colin Brauner, Greg Goss and Steve McCormick

Abstract: There have been many recent advances in both our understanding of fish ion transport mechanisms and acid-base physiology and how they are regulated in fish. This symposium will bring together top researchers in the broad field of ion and of acid-base regulation in fishes to discuss current topics and future directions for the field. We plan to have sub-sections within the symposium that will deal with new advances in our understanding of: a) endocrine and mechanistic control of ion regulation, b) smoltification, and c) acid-base regulation, both in relation to basic and applied research including climate change and conservation physiology. The symposia will highlight researchers using whole animal approaches and those following more cellular and molecular mechanisms underlying these regulatory events. This symposium will bring together speakers interested in a holistic approach to understanding the mechanisms involved in alteration and regulation of ion and acid-base status in fishes and the implications of this for whole animal function and responses to altered environments.

IONS I Ion and acid-base regulation in fishes

Differential gene expression analysis of zebrafish gills from fish exposed to low pH, low ionic strength waters

Goss, Greg (*ggoss@ualberta.ca*) Oral Presentation
Co-Author: Kovac, Anthony
University of Alberta

Abstract: Na+/H+ exchangers (NHE) have been shown to mediate Na+ ionoregulatory status in some model fish species living in circumneutral pH and [Na+] > 1 mM freshwaters, very low pH water (< pH 5) coupled with low ionic strength conditions (< 500 uM), places severe thermodynamic constraints on NHE function. We recently described the recovery of Na+ uptake in zebrafish exposed to pH 4.0 water by a novel K+-driven Na+ uptake mechanism whereby Na+ uptake increases in parallel with increased K+ efflux after 8 h of pH 4.0 exposure (Clifford et al. 2022). To follow up, we contructed gill transcriptomes from zebrafish exposed to either control for pH 4 for 4 or 8 hours.~ 30 candidate genes involved in zebrafish Na+ homeostatic response to low pH freshwaterwere established using differential expression of gene (DEG) analysis and validation through quantitative polymerase chain reaction (qPCR) analysis. However, only a single protein with transmembrane functionality was identified as increasing in expression in both 4 and 8 h periods. Synaptic vesicle 2 protein-like (svopl) was found to be significantly upregulated in zebrafish gill tissue. Svopl has predicted functionality as an ion transporter with 12 transmembrane domains but experimental studies and information on Svopl and its human orthologs are absent in the literature. In summary, we have identified Svopl as a

valid candidate gene for the Na+ homeostatic response in zebrafish to low pH freshwater and await further characterization of expression and localization.

IONS I Ion and acid-base regulation in fishes

What are the ecophysiological implications of periodic acidification of riverine systems?

Zimmer, Alex (*alex.zimmer@unb.ca*) Oral Presentation
University of New Brunswick

Abstract: In the 1980s, the acid rain crisis that led to widespread fish die-offs in many regions was the impetus for a large body of research on the effects of hydrogen ions on fish physiology. At severely depressed pH levels (< 4.0), acid-sensitive freshwater fishes experience a net loss of Na+ and Cl- resulting from the inhibition of active ion uptake and a concomitant stimulation of passive ion loss. While the effects of severe, anthropogenic acidification by inorganic acids have been well-characterized, far less is known regarding the effects of natural acidification processes driven by organic acids. Many freshwater systems with low acid-neutralizing capacity are subject to natural acidification events throughout the year. Acidification linked to spring snowmelt events is a well-described phenomenon in many systems, and work by our group has additionally gathered preliminary evidence suggesting that large rainfall events can also cause acidification. In both instances, acidification can result in pH values that are known to elicit ionoregulatory disturbances in laboratory experiments. In this talk, I will describe the nature of periodic acidification events in freshwater systems, including our work on rivers in New Brunswick, Canada, highlight the link between ionoregulatory physiology and acid-sensitivity in freshwater fishes, and discuss the potential ecophysiological implications of periodic acidification events in freshwater rivers. While pH has been highlighted in previous studies as a factor that shapes species distribution patterns in permanently acidic lakes, the effects of periodic acidification events on fish assemblages and on fish physiology have received far less attention.

IONS I Ion and acid-base regulation in fishes

Gill methylome shifts across multigenerational acidification in marine medaka

Tseng, Yung-Che[1] (*yctseng@gate.sinica.edu.tw*) Oral Presentation
Co-Author: Liu, Tzu-Yen[1]; Nash, Mikyla Tara[1]; Allen, Garett Joseph Patrick[2]
[1]Academia Sinica, [2]Acadia University

Abstract: Epigenetic modifications may facilitate the acclimatization of marine fishes to ocean acidification over repeated generations. Prolonged CO_2 exposure in marine medaka (Oryzias melastigma) embryos produces hypomethylation of ion transport genes; however, it is unclear if analogous reprogramming happens in adult osmoregulatory organs. We analyzed genome-wide DNA methylation in the gills of medaka maintained at pH 7.6 during

three successive generations (F0, F1, F2) utilizing Nanopore sequencing. Wild-type gills displayed the largest counts of methylated CpG sites, whereas acidified generations demonstrated a gradual demethylation at transcription start sites. Genes with differential methylation were enriched in post-transcriptional regulatory mechanisms and cytoskeletal architecture. Simultaneous transcriptome analysis demonstrated a concomitant increase of metabolic and protein localization pathways in subsequent generations. Demethylation patterns at crucial regulatory loci were comparable between embryonic and adult gill tissues, indicating conserved epigenetic targets throughout ontogeny stages. The data suggest that intergenerational acclimatization entails synchronized methylation alterations at upstream regulatory sites, likely enhancing downstream physiological responses to prolonged acidity.

IONS I Ion and acid-base regulation in fishes

Linking Molecular Transport Processes to Acid–Base Regulation Strategies in Fishes

Shartau, Ryan[1] (*ryan.shartau@wsu.edu*) Oral Presentation
Co-Authors: Kostelnik, Magaleate[1] and Flowers, Dillon[2]
[1]Washington State University, [2]Oklahoma State University

Abstract: Fishes exhibit two primary strategies of acid–base regulation: coupled pH regulation (CPR), in which extracellular pH is maintained to support intracellular pH (pHi), and preferential pHi regulation (PPR), in which tissues maintain pHi independently of changes in blood pH. While these patterns are well characterized at the organismal level, the cellular and molecular mechanisms that distinguish them remain poorly understood. To address this gap, we examined species representing each strategy, rainbow trout (Oncorhynchus mykiss; CPR) and channel catfish (Ictalurus punctatus; PPR), to test whether differences in pHi regulation are associated with conserved ion transport pathways. Fish were exposed to elevated CO_2 to induce acid–base disturbance, and tissue-specific responses were examined across multiple organs. We targeted members of theslc4family of bicarbonate transporters, integrating gene expression and protein-level analyses to identify candidate mechanisms underlying pHi regulation. Preliminary results indicate distinct, tissue-specific responses between species consistent with differences in pHi regulatory strategy, implicating bicarbonate transport processes as a potential mechanism supporting PPR. Ongoing RNA-seq analyses are expanding these findings to a systems-level understanding of acid–base regulation in heart and liver tissues. Future research will examine how these mechanisms emerge during development and vary among tissues, testing the hypothesis that intracellular pH regulation is most robust early in ontogeny. By linking molecular transport processes to organismal strategies, this work advances a framework for predicting how fishes respond to environmental stressors, including elevated CO_2, warming environments, and changing water chemistry.

IONS I Ion and acid-base regulation in fishes

Distinct acid-base regulatory strategies in teleosts and elasmobranchs under ocean acidification

Chuang, Hsin-Ju[1] (*ruby840504@gmail.com*) Oral Presentation
Co-Author: Hyodo, Susumu [1]; Hwang, Pung-Pung [2]
[1] The University of Tokyo, [2]Academia Sinica

Abstract: Ocean acidification (OA), caused by excess anthropogenic carbon dioxide (CO2) emission, poses a significant threat to marine ecosystems. In recent years, the effects of OA on teleosts have been widely investigated, greatly advancing our understanding of their acid-base regulatory mechanisms. In contrast, our knowledge of how elasmobranchs regulate acid-base balance under OA remains limited. Some studies suggested that elasmobranchs may respond to elevated CO2in ways similar to teleosts. However, elasmobranchs are ureotelic, possess distinct ionocytes subtypes in their gills for acid-base regulation, and primarily secrete ions through the rectal gland, indicating potentially different physiological strategies. Using cloudy catshark (Scyliorhinus torazame) as a model species, this study compares the physiological responses of teleosts and elasmobranchs to CO2-induced environmental acidification and aims to elucidate the acid-base regulatory mechanisms of ionocytes in elasmobranch gills. When exposed to OA conditions, plasma pH in catsharks increased rapidly and returned to level comparable to the control within 24 hours of exposure. The activation of A- and B- type ionocytes in the gills may contribute to restoring acid-base balance, and coordination between these cell types likely play an important role in maintaining internal homeostasis. Compared with the well-studied ionocytes of teleosts, elasmobranch ionocyte subtypes appear to exhibit a distinct regulatory machinery for acid-base balance. Understanding these physiological mechanisms provides insight into how different groups of fishes may cope with an acidifying ocean. Integrating physiological responses with ecological perspectives will improve predictions of species resilience and vulnerability under ongoing ocean acidification.

IONS I Ion and acid-base regulation in fishes

Characterizing bicarbonate transporters and their osmoregulatory roles in the Sea Lamprey intestine

Flowers, Dillon (*dillon.flowers@okstate.edu*) Oral Presentation
Co-Author: Shaughnessy, Ciaran
Oklahoma State University

Abstract: An important aspect of osmoregulation in marine fishes is intestinal ion transport, which facilitates water absorption in proximal intestine, calcium carbonate (CaCO3) formation in distal regions, and maintaining acid-base balance during hypercapnic exposure. However, the molecular mechanisms of intestinal ion and water absorption, CaCO3 precipitation, and acid-base balance are not well understood in lampreys, the most basal vertebrate lineage capable of osmoregulation. Here, we present a

molecular and functional characterization of HCO3--transporting solute carrier (Slc4) proteins in the lamprey intestine. Our analysis of the Sea Lamprey (Petromyzon marinus) genome revealed a limited and structurally related set of Na+-HCO3--cotransporter (Nbc)-like and 2 Cl-/HCO3--exchanger (Ae)-like Slc4 family members, compared to the diverse and distinct set of gnathostomes Slc4 classes. To investigate whether lamprey utilize these transporters in intestinal ion transport, we analyzed a tissue distribution of their transcriptional expression in seawater (SW)-acclimated juvenile Sea Lamprey. Next, we analyzed gene expression across regions of the intestine (proximal to distal), life stages (larvae and juvenile), and salinities (ion poor to hypersaline). We found prominent intestinal expression of Ae-like genes within the anterior and posterior intestine and Nbc-like genes within the esophagus and anterior intestine, the latter of which increased in expression during metamorphosis and salinity acclimation. Together, these findings preliminarily suggest ion–$HCO3^-$ transport by slc-like genes plays an important role in osmoregulation in the Sea Lamprey intestinal tract. Further investigation of transporter regulation is needed to better understand their role in the lamprey gastrointestinal tract during seawater adaptation.

IONS I Ion and acid-base regulation in fishes

Up-regulation of Fgf23 contributes to kidney stone formation in stc1a-/- mutant fish

Aburatani, Naotaka[1] (*naotakaa@umich.edu*) Oral Presentation
Co-Author: Peterson, Brian[2] and Duan, Cunming[1]
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Abstract: Nephrocalcinosis (kidney stones) presents a significant challenge in farmed Atlantic salmon smolt, yet its underlying mechanisms and diagnostic markers remain poorly understood. Previously, we reported that genetic deletion of Stanniocalcin1a (Stc1a) in zebrafish leads to the formation of calcified deposits within the kidney. However, the chemical composition and molecular basis of these deposits were unclear. In this study, we demonstrate that these renal deposits consist primarily of calcium and phosphate, forming within the proximal convoluted tubule of the larval nephron. Morphological analysis and in situ hybridization using renal marker genes revealed no major structural defects in stc1a mutants. Gene expression profiling showed a pronounced upregulation of fibroblast growth factor 23 (fgf23), a hypophosphatemic hormone, while expression of renal phosphate transporters slc20a1a and slc34a1a, known downstream targets of Fgf23, were downregulated. These molecular changes were abolished when stc1a mutant larvae were reared in low Ca2+ conditions, suggesting that hypercalcemia resulting from loss of Stc1a induces fgf23 expression and disrupts phosphate reabsorption, leading to kidney stone formation. Supporting this hypothesis, CRISPR-Cas9-mediated knockout of fgf23 partially rescued the kidney stone phenotype in stc1a mutants, and pharmacological inhibition of Fgf23 signaling similarly reduced stone occurrence. Our findings indicate that loss of Stc1a and consequent up-regulation of Fgf23 contribute to kidney stone formation in zebrafish. Furthermore, aberrant expression of Stc1a and Fgf23 may serve as potential diagnostic markers for nephrocalcinosis in fish.

IONS I Ion and acid-base regulation in fishes

Gill remodelling and increased Rh glycoprotein abundance enhance postprandial ammonia excretion by blood-feeding parasitic juvenile sea lamprey.

Wilkie, Michael (*mwilkie@wlu.ca*) Oral Presentation
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Abstract: Sea lamprey (Petromyzon marinus) are an anadromous jawless fish that spend the early part of their life cycle burrowed in streams as filter-feeding larvae before completing a complex metamorphosis into parasitic blood-feeding juveniles, which migrate downstream to the sea or the Great Lakes where they prey on teleost fishes. Blood ingestion by juvenile sea lamprey leads to high rates of protein catabolism, which generates large amounts of ammonia. We previously demonstrated that juvenile sea lamprey have a high capacity to excrete ammonia following blood ingestion, with postprandial ammonia excretion rates (JAmm) approaching 2500 mmol kg-1h-1, up to 25-fold greater than basal rates, and likely mediated by Rh glycoproteins (Rhg). Rhg's facilitate NH3 diffusion across gills, and are most abundant in seawater (SW) ionocytes in juvenile sea lampreys. Adjacent lamellae contain intercalated ionocytes, which possess acid excreting H+-ATPase's, that may trap excreted NH3 as NH4+ to sustain JAmm across the gills. We tested the hypothesis that Rhg and H+-ATPase protein expression increased after feeding to promote postprandial Rhg-mediated JAmm following blood ingestion. The juvenile sea lamprey fed on rainbow trout for 3 weeks, under carefully controlled conditions, after which postprandial JAmm, branchial Rhg and H+-ATPase protein expression, and gill morphometrics were quantified. Postprandial JAmm was 13-fold greater than basal rates, and accompanied by a 3-fold increase in Rhg expression, with no change in H+-ATPase abundance. Notably, lamellar length increased, potentially promoting greater H+-excretion capacity and NH3 -trapping. We conclude that greater Rhg protein expression and gill remodeling enhance postprandial ammonia excretion by juvenile sea lamprey.

IONS I Ion and acid-base regulation in fishes

Possible ammonia removal mechanism in the seahorse brood pouch revealed by rhesus glycoprotein expression and localization

Kawaguchi, Mari[1] (*k-mari@sophia.ac.jp*) Oral Presentation
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Abstract: In seahorses (family Syngnathidae), the male brood pouch provides a protected environment for developing embryos. Because the pouch remains closed during pregnancy, nitrogenous waste excreted by the embryos must be removed through a

specialized physiological mechanism. This study investigated the expression and localization of rhesus (Rh) glycoproteins—putative ammonia transporters—in the brood pouch of the pot-bellied seahorse Hippocampus abdominalis. Expression of the RhAG, RhBG, and RhCG2 genes in the pouch was confirmed by RT-PCR. In situ hybridization and immunohistochemistry further revealed that RhBG and RhCG2 are localized in the inner epithelial layer of the pouch, whereas RhAG is present in erythrocytes and in the endothelium of blood vessels. Based on these findings, we propose the following ammonia elimination pathway. Ammonia released by the embryos is transported from the pouch lumen across the inner epithelium via apical RhCG2 and basolateral RhBG, entering the connective tissue beneath the epithelium. There, ammonia permeates RhAG-expressing endothelial cells into the vascular lumen and is subsequently taken up by RhAG-expressing erythrocytes, which carry it to the gills through the bloodstream. Finally, ammonia is released from erythrocytes via RhAG-expressing pillar cells and RhBG-/RhCG2-expressing pavement cells, and is excreted into the surrounding seawater. This study provides the first molecular evidence for a specialized waste-removal system within the seahorse brood pouch, highlighting a unique physiological adaptation associated with male pregnancy in syngnathid fishes.

IONS I Ion and acid-base regulation in fishes

Integrating gene expression, eQTL, and population genomic variation to reveal molecular adaptation in prickly sculpin (Cottus asper) across British Columbia

Liu, Shuang (*liu.shuang@ubc.ca*) Oral Presentation
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Abstract: Understanding the genomic and transcriptomic mechanisms underlying local adaptation is a central theme in evolutionary biology. The prickly sculpin (Cottus asper), a euryhaline fish distributed across freshwater and brackish habitats in northwestern North America, provides an ideal system to study adaptive divergence. Previous work identified local adaptation in ion regulation across distinct life-history ecotypes in British Columbia, but the molecular mechanisms, e.g., genetic variation and gene regulation, underlying such local adaptation remain unclear.

We integrated population genomics, gene expression profiling, and expression quantitative trait locus (eQTL) mapping using gill RNA-seq data from populations occupying three distinct habitats. Differential expression analyses revealed extensive transcriptional divergence among regions and region-by-salinity interactions. Gene ontology enrichment indicated functions related to ion transport, hypoxia response, metabolism, hormone signalling, and immune processes. Cis-eQTL analyses identified modifier-impact SNPs significantly associated with expression variation, yielding 33,962 SNP–gene associations linked to regional differences and 1,265 associated with genotype-by-environment interactions. Integrating eQTLs with allele-specific expression (ASE) further refined candidate cis-regulatory variants, identifying 6,372 ASE modifier SNPs associated with

habitat-related DE genes and 172 linked to interaction effects. Overlap of FST outlier loci with cis-regulatory regions of DE genes identified candidate adaptive variants affecting genes associated with ion transport, hypoxia response, signalling, metabolic, and immune pathways, supporting local selection on regulatory variation.
Together, our results show that cis-regulatory variation drives gene expression divergence and local adaptation in ecologically relevant pathways related to ion balance and environmental stress, clarifying the molecular architecture of environmental adaptation across heterogeneous habitats.

IONS I Ion and acid-base regulation in fishes

Salinity and sex dependent ionoregulatory mechanisms in reproductively active Fundulus heteroclitus

Diakanastasis, Eleni[1] (*elenid@my.yorku.ca*) Oral Presentation
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Abstract: This study considered the idea of sex specific differences in the ionoregulatory physiology of reproductively active male and female mummichogs (Fundulus heteroclitus). During the breeding season, male and female fish exhibiting gonadosomatic indices of 2.49 ± 0.203 and 12.54± 1.48 respectively, were held in either freshwater (FW, 0‰), brackish water (BW, 15‰), or seawater (SW, 30‰) for 14 days. Following this, systemic as well as organ-specific endpoints of salt and water balance were examined.
Muscle moisture content did not differ across sexes or salinity treatments. Muscle and carcass chloride concentrations were also similar between sexes within each salinity; however, both sexes exhibited elevated chloride levels in BW and SW relative to FW. Similarly, carcass and muscle sodium concentrations increased progressively from FW to SW. Salinity did not significantly alter gill Na^+/K^+-ATPase activity in female fish, whereas Na^+/K^+-ATPase activity increased significantly in SW acclimated male fish. Despite this pattern, no overall sex specific differences were detected in gill Na^+/K^+-ATPase activity. Analysis of transcript abundance revealed sex specific regulation of pathways associated with both paracellular and transcellular ion transport. Several transcripts related to epithelial barrier function and ion transport displayed sex dependent patterns of abundance across salinity treatments. Together, these findings suggest that male and female euryhaline fish employ partially distinct physiological strategies to cope with changes in environmental salinity.

IONS I Ion and acid-base regulation in fishes

Hormonal control of determinants of hydromineral balance in gill and intestine of Atlantic killifish

Tipsmark, Christian (*tipsmark@uark.edu*) Oral Presentation
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University of Arkansas

Abstract: The coordinated control of compensatory branchial ion transport and intestinal water handling is critical for maintenance of hydromineral balance in teleost fish. Among the hormones regulating phenotypic plasticity, prolactin, growth hormone and cortisol were the focus of these studies, using the study species, Atlantic killifish (Fundulus heteroclitus).With a combination of in-vivo injection experiments and ex-vivo explant experiments we examined the role of these hormones in control of expression of determinants of gill and intestinal ion and water transport. In the gill, gene expression central to salt secretion was stimulated by cortisol and generally dampened by prolactin (cftr, nkcc1, cln10 paralogs) while proteins critical in fresh water were stimulated by prolactin (aqp3, ncc2, cldn30c, cldn32a). Importantly, a synergistic effect of prolactin and cortisol was observed on freshwater target genes (aqp3, ncc2b) emphasizing the dependence of prolactin on cortisol in its mode of action. In the intestine prolactin consistently downregulated expression of proteins associated with the seawater phenotype, including bicarbonate transporters (slc26a3a and slc4a4a) and nkcc2. Cortisol, on the other hand, stimulated transcript abundance of both intestinal bicarbonate transporters and ion/water transporters (cftr, nkcc2, aqp8). These studies emphasized the interdependence of prolactin on cortisol for mode of action in gill and furthermore surveyed the role of the examined osmoregulatory hormones in the intestine. Positive controls confirmed that growth hormone acts in our experimental system and we will present data related to the role of this hormone in the context of hydromineral homeostasis, and gill and intestinal function.

IONS I Ion and acid-base regulation in fishes

Prolactin, cortisol, and extracellular osmolality regulate cftr, ostf1, and sgk1 in tilapia ionocytes

Breves, Jason[1] (*jbreves@skidmore.edu*) Oral Presentation
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Abstract: Euryhaline fishes can inhabit salinities ranging from fresh water (FW) to seawater (SW) because their endocrine system adaptively matches the ion-transporting capacities of branchial ionocytes to the environment. Ionocytes also directly sense extracellular osmolality and adjust their functions accordingly; therefore, we investigated the interplay between osmotic conditions and osmoregulatory hormones in Mozambique tilapia (Oreochromis mossambicus) to further elucidate the bases of euryhalinity. We investigated whether prolactin (Prl) supports FW acclimation by counteracting osmotic and hormonal

signals that initiate branchial responses to SW. Initially, we combined hypophysectomy, hormone replacement, and RNA-Seq to identify cystic fibrosis transmembrane conductance regulator (cftr), osmotic stress transcription factor 1 (ostf1), and serum- and glucocorticoid-inducible kinase 1 (sgk1) as Prl-repressed genes. We then characterized their in vivo and in vitro regulation by environmental salinity, Prl, cortisol, and extracellular osmolality. Our findings indicate that 1) Prl's capacity to antagonize cortisol-stimulated cftr expression depends on extracellular osmotic conditions; 2) Prl and cortisol play opposing roles in regulating branchial ostf1 expression; and 3) Sgk1 is expressed in 'SW-type' ionocytes, where interactions among Prl, cortisol, and osmotic conditions influence its expression. Ultimately, the combined actions of osmotic stimuli, Prl, and cortisol shape the branchial expression of cftr, ostf1, and sgk1. While hyperosmotic conditions and cortisol promote activation of these genes during SW acclimation, Prl supports FW adaptation by suppressing these genes and promoting processes underlying active ion uptake. Thus, euryhaline tilapia adjust the ion-transporting activity of their branchial ionocytes by integrating multiple regulatory cues.

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Functional organization of ion transport in the trunk skin of European seabass (Dicentrarchus labrax): Evidence of an apical localization of Na+/K+-ATPase

Sundh, Henrik[1] (*henrik.sundh@bioenv.gu.se*) Oral Presentation
Co-Author: Pinto, Patricia [2]; Thorne, Michael [3]; Sundell, Kristina[1]; Guerreiro, Pedro M.[4] and Power, Deborah[2]
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Abstract: Fish trunk skin is regarded as a passive diffusion barrier that is essential for homeostasis. Conventional knowledge of transporting epithelia places Na+,K+-ATPase (NKA) to the basolateral membrane (inside) such as in the vertebrate skin, kidney, bladder, gut and the fish gill. Herein, we show that the European sea bass (Dicentrarchus labrax) trunk skin develops a positive transepithelial electrical potential (TEP) and a corresponding short-circuit current (SCC) after mounting in Ussing-chambers at identical Ringer´s solution conditions indicating active transport. A regional difference was found where the skin in the dorso-anterior region of the fish displayed a higher TEP and SCC. Immunohistochemistry revealed NKA, NKCC and CFTR positive cells in the epidermis. Interestingly, ouabain instantly inhibited the TEP and SCC when added to the epidermal/apical (out)side while no effects on TEP or SCC were obtained when ouabain was added to the dermal/basal (in)side. The location of functional tight junctions was indirectly determined by adding biotin to the dermal side for 60 min. Immunostaining of biotin revealed that the protein penetrated the dermis and reached all the way up to the basal layer of the epidermis, but it did not penetrate the epidermis. Collectively our data provide evidence for an unconventional organization of the seabass skin where the NKA appears to be localized to the apical membrane while the tight junctions appear to be localized to the deeper epidermal layer close to the basal membrane. This suggested

"inside-out" configuration of the sea bass skin is novel and challenges conventional understanding of epithelial transport.

IONS I Ion and acid-base regulation in fishes

Photoperiod- and salinity-dependent transcriptional regulation of renal CaSR and calcium transporters supports calcium homeostasis during seawater acclimation in Atlantic salmon

Takvam, Marius (*m.takvam@uib.no*) Oral Presentation
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Abstract: In teleost fishes, the kidney plays a central role in maintaining divalent ion homeostasis in both freshwater (FW) and seawater (SW). While the excretion of Mg^{2+}, Ca^{2+} and SO_4^{2-} in euryhaline teleosts is widely assumed to occur via tubular secretion, this mechanism remains largely inferred from urine composition and has been only sparsely investigated at the molecular level. Here we investigated whether smoltification and subsequent salinity exposure transcriptionally regulate renal Ca^{2+} sensing and transport pathways to stabilize systemic Ca2+ during SW acclimation in Atlantic salmon. In silico analyses identified an expanded repertoire of calcium transporters within the NCX, PMCA, and TRPV5 family. Despite conservation of core catalytic and ion-binding domains, salmonid-specific substitutions were identified in predicted regulatory domains, potentially conferring paralog-specific sensitivity to environmental cues. Renal casr, ncx1b1 and pmca1b1 mRNA levels displayed a transient increase during smoltification and followed a second increase in seawater. Most of the remaining pmca and ncx paralogs were upregulated following 7 days in SW. These transcriptional responses coincided with a short-term increase in plasma Ca^{2+} following 7 days SW exposure that normalized after 30 days. Concurrently, urinary Ca^{2+} concentrations of seawater acclimated salmon were approximately four times the values of freshwater, consistent with a functional shift from renal Ca^{2+} reabsorption to secretion. Together, these findings suggest photoperiod- and salinity-dependent upregulation of renal CaSR and Ca2+ transporters in Atlantic salmon. Our findings provide new insight into the transcriptional mechanisms underlying renal Ca^{2+} sensing and handling in euryhaline teleosts.

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Impact of commercial feed mineral load on urine composition and transcriptional regulation of divalent ion transporters in Atlantic salmon kidney

Denker, Elsa[1] (*elsa.denker@uib.no*) Oral Presentation
Co-Author: Le, Thi Hong Gam[1]; Takvam, Marius[1]; Mullins, Julia[2]; Knusten, Markus[1]; Karlsen, Rita[1] and Nilsen, Tom Ole[1]
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Abstract: While it is established that teleosts in freshwater need to reabsorb divalent ions from the water through the kidney, it has been shown that this paradigm can be shifted depending on the feed status of the fish, especially if commercial feed is involved. Time series on urine collected in rainbow trout have shown that the introduction of commercial pellets in unfed fish impacts kidney divalent ion handling, increasing net excretion of Ca2+ and Mg2+. To unravel the molecular mechanisms underlying these changes, we studied the expression of Ca2+, Mg2+ and Pi transporters in the kidney of Atlantic salmon fed three diets containing increasing mineral load, specifically for Ca, Mg, and P. After acclimation, plasma, spot-sampled urine and tissue samples were collected in all groups, then feeding was stopped for 72h while urine composition was monitored through urinary bladder catheterization, before terminal sampling. We observed that the transporters known to be involved in Ca2+, Mg2+, and Pi secretion during freshwater to seawater transition in juvenile Atlantic salmon such as pmca1a2, ncx2a, ncx4b, slc41a1-1,or slc34a1a2 were down-regulated after 72h starving, in all three diet groups, indicating that even under a mineral load lower than average commercial feed, fish increased net excretion of Ca2+, Mg2+, and Pi. These changes were correlated with urine composition changes. This finding has implications not only for experimental studies but also for commercial aquaculture, as increased renal excretion of Ca^{2+} and Mg^{2+} may elevate the risk of supersaturation events ultimately leading to kidney stone formation.

IONS I Ion and acid-base regulation in fishes

The calcium binding protein S100a11 regulates zebrafish ionocyte quiescence–proliferation balance

Shih, Shang-Wu (*swshih@umich.edu*) Oral Presentation
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Abstract: Reactivation of quiescent cells is essential for maintaining tissue homeostasis in response to stress and injury. However, the mechanisms controlling the proliferation-quiescence decision are not fully understood. We recently demonstrated that Na+/K+ ATPase-rich cells (NaR cells), a specialized ionocyte responsible for calcium uptake in zebrafish, enter a quiescent state upon expression of the constitutive calcium channel Trpv6. Genetic deletion of Trpv6 or reduction of extracellular Ca^{2+} concentration decreased cytoplasmic free Ca^{2+} ($[Ca^{2+}]c$) and reactivated NaR cells. This reactivation was inhibited by chelation of $[Ca^{2+}]c$, highlighting the importance of cytoplasmic calcium in the process.

The regulatory mechanisms underlying [Ca^{2+}]c remain unclear.In this study, we identified S100a11, a member of the S100-family of Ca^{2+}-binding proteins, as being exclusively expressed in NaR cells. Analysis of available single-cell RNA sequencing datasets revealed strong enrichment of s100a11 transcripts in both adult and larval NaR cells, which was confirmed using fluorescence in situ hybridization. F0 CRISPR knockout of S100a11 abolished low Ca^{2+}-induced NaR cell reactivation and proliferation, while had no impact on NaR cell numbers under normal calcium conditions.To further explore the role of S100a11, we examined stc1a-/- mutants, which exhibit increased NaR cell numbers due to activation of the Igf-Akt-Tor signaling pathway. F0 S100a11 knockout impaired this elevated NaR proliferation.Collectively, these results establish s100a11 as a novel marker for NaR cells and reveal a role for S100a11 in regulating the quiescence-proliferation balance in these cells. These findings provide a foundation for future studies investigating the interplay between intracellular calcium dynamics and Igf-Akt-Tor signaling.

IONS I Ion and acid-base regulation in fishes

Genomic variation that distinguishes euryhaline from stenohaline Fundulus species

Bernat, Tatum[1] (*tsbernat@ucdavis.edu*) Poster Presentation
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Abstract: Evolved differences in osmoregulatory physiology have allowed fishes to colonize vastly different habitats and much remains to be learned about the evolved genomic variation that underlies the array of osmoregulatory strategies. In Fundulus killifish, euryhaline lineages have repeatedly and independently radiated from marine into freshwater environments, accompanied by a loss of euryhalinity and a gain of freshwater specialization. These repeated evolutionary events provide a unique opportunity to apply comparative genomic methods to investigate the variation across several species, while accounting for phylogenetic relatedness. We hypothesized that the loss of euryhalinity would manifest as patterns of protein sequence or protein family evolution that differ between euryhaline and stenohaline killifish; if specialized protein function is important for euryhalinity but not for stenohalinity, then we predict that those protein sequences will be less conserved in derived freshwater species. Additionally, proteins important for derived freshwater specialization should show accelerated evolutionary rates due to directional selection. Gene family expansion or contraction may also underlie loss or euryhalinity or gain of freshwater specialization. We found divergent rates of gene family evolution and protein sequence evolution that offer insight into the mechanisms that support both osmoregulatory flexibility and freshwater specialization. Complementary to these findings, we are also pursuing comparative gill transcriptomics and comparative physiology analyses to further synthesize our functional understanding of diverse osmoregulatory physiologies and how they evolve, integrating inference from genomic variation, genome function, and physiological responses.

IONS I Ion and acid-base regulation in fishes

Gill ionocyte diversity and metabolic adaptation to freshwater in Indian medaka (Oryzias melastigma)

Guh, Ying-Jey (*yingjeyguh@gmail.com*) Poster Presentation
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Academia Sinica

Abstract: Freshwater (FW) environments fluctuate in ionic composition, nutrient availability, and oxygen tension, likely shaping the diversity of branchial ionocyte taxonomy. Using single-cell transcriptomics and trajectory analysis of gill epithelial cells from marine medaka (Oryzias melastigma) acclimated to freshwater and seawater, we revisited ionocyte classification and found that FW-specific NCC (Na+-Cl- cotransporter) ionocytes diverge from the same ionocyte precursor lineage as NHE (Na+-H+ exchanger) ionocytes, indicating that NCC cells represent a FW-specialized branch of a shared developmental program. This specialization is accompanied by metabolic reprogramming. Consistent with reports that NADPH-producing routes track dominant metabolite flux, FW-NCC cells show elevated hk2 expression and increased pentose phosphate pathway (PPP) signatures, supporting a glucose-forward program that supplies cytosolic NADPH for redox buffering. In contrast, NHE ionocytes preferentially express mitochondrial NADPH enzymes (idh2, nnt), suggesting a distinct NADPH strategy and fuel-redox coupling more compatible with lipid-supported metabolism. We propose that metabolic diversification among ionocyte subtypes increases gill plasticity, enabling fishes to match energetic and antioxidant strategies to fluctuating FW constraints. In particular, glucose preference in NCC cells may enhance fuel-use flexibility in nutrient-poor and hypoxia-prone FW conditions, contributing to osmoregulatory resilience during freshwater invasion.

IONS I Ion and acid-base regulation in fishes

Phytogenic modulation of the cortisol mediated stress response and branchial Na^+/K^+-ATPase activity in European seabass

Sara Cartan[1] Poster Presentation
Co-Author: Mancera Romero, Juan Miguel[1], Jerez-Cepa, Ismael[1] and Martínez Rodríguez, Gonza[2]
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Abstract: Live fish transport is a critical event in aquaculture, often triggering a stress response that disrupts homeostasis and can lead to hydromineral imbalance by altering branchial Na^+/K^+-ATPase (NKA) activity. While the search for natural sedatives is essential for fish welfare, their addition to the water may also present osmoregulatory challenges. This study evaluated the efficacy of a phytogenic aqueous product (FEC) in modulating the stress response and NKA activity in European seabass (Dicentrarchus labrax) following 1, 3, and 6 h of transport. An initial 1 h transport simulation using 2, 10, and 20 µL L^{-1} of the

FEC sedative identified 10 μL L^{-1} as the optimal concentration for reducing brain cortisol receptor expression (nr3c1 and nr3c2). Moreover, its application did not impair NKA activity or hydromineral balance after 1 h of transport. The use of the FEC sedative during the 3- and 6-h transports effectively modulated the stress response, improved recovery (24 h post-transport), and prevented the significant disruption of NKA activity observed after 6 h of transport and subsequent recovery. Multiple correlation analysis showed that NKA activity was negatively correlated with plasma cortisol, glucose, and amino acid levels, suggesting a link between ionoregulatory stability and energy availability. Consequently, by modulating the cortisol-mediated response, this sedative helps preserve branchial NKA efficiency and contributes to osmotic balance. These findings highlight FEC as a viable strategy to enhance resilience and maintain osmoregulatory integrity in seabass during aquaculture transports.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Applying knowledge of fish biology to sustainable aquaculture

Andrea Frommel and Tillmann Benfey

Abstract: Although fish farming has been practised for thousands of years, it is only in the last few decades that global fish production from aquaculture has outpaced capture fisheries as a source of food for human consumption and for conservation of threatened populations. This recent and rapid transition from hunting and gathering wild fish to farming them has had many positives but also raises concerns about sustainability that are being compounded by the effects of climate change on the freshwater and coastal marine environments historically used for aquaculture. The goal of this symposium is to bring together speakers with shared interests in applying fundamental knowledge of fish biology to ensuring the continued responsible growth of aquaculture and hatchery production that considers the welfare of the animals and the environmental impacts of their rearing systems. Possible topics include genetics (selective breeding, gene editing, ploidy manipulation), nutrition (alternative feed ingredients and immunostimulants), health (novel vaccines, microbiome, probiotics), environmental physiology (realizing phenotypic plasticity in hyperthermia and hypoxia tolerance), practical welfare indices, recirculating aquaculture systems and hatchery practices.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Impact of intergenerational enrichment on juvenile westslope cutthroat trout methylome and phenotypic traits

Firth, Britney (*blfirth@ualberta.ca*) Oral Presentation
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Abstract: Hatcheries are used worldwide to supplement game fish such as salmonids. However, fish from barren hatchery environments can have reduced fitness and adversely affect wild individuals. Enrichment (i.e., substrate, shelter, plants) can minimize the environmental mismatch between hatchery and wild phenotypic traits likely through epigenetic mechanisms such as DNA methylation. DNA methylation signatures can alter gene expression and are often passed from parent to offspring. Enrichment at multiple life stages is labour intensive and impractical in large production hatcheries. Therefore, it is important to understand the relative benefits of parental (i.e., epigenetic inheritance) versus offspring (i.e., acclimation) enrichment on DNA methylation signatures and phenotypic traits. To assess the effect of parental versus offspring enrichment, westslope cutthroat trout (Oncorhynchus clarkii lewisi) were split between barren and enriched environments, bred, and offspring were split between the same treatments. Enzymatic methyl-seq was used to characterize the methylome of whole alevin and fry from all

treatments. To determine if methylome changes impact phenotypic traits relevant for post-release survival, fry anti-predator behaviour and metabolic rate were assessed. These data will characterize the life-stage specific influences of environmental enrichment and define the relative importance of parental versus offspring enrichment on hatchery-induced phenotypes, with implications for post-release of captive trout.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Balancing natural and artificial selection in captive rearing programs

Maynard, Desmond (*desmondmaynard@msn.com*) Oral Presentation

Abstract: Imperiled populations threatened with extinction are sometimes taken into captive rearing and breeding programs to preserve their unique suite of genetic traits adapting them to their natural local environment.These traits include morphological, physiological, life history and behavioral adaptations that differ between populations.Conservation programs must carefully consider adopting husbandry approaches designed to maintain these unique local adaptions during culture.Otherwise, misguided or unintended artificial selection may shift naturally balanced polymorphisms in directions that lower fitness when populations are returned to their natural environment. In Pacific salmon populations reproductive traits of concern include age of maturation, return time, spawn time, nest site characteristics, substrate preference, nest construction, nest defense, egg size and fecundity. Salmonid culture tools with the potential to counter these reproductive concerns include spawning channels and sourcing each generation as eyed eggs, parr, or smolts. Survival traits of concern include emergence timing, habitat selection, habitat use, freshwater habitat migration, anadromous migration, foraging behavior, digestive physiology and physiological tolerances (temperature, salinity, dissolved oxygen, etc.). Culture technology using locally adapted temperature profiles, native area photoperiods, more natural diets, seawater rearing during ocean life history phases, and timing life stage events to match natural profiles may help maintain the natural balance of alternative traits in the cultured population. It is anticipated the success of using artificial culture to restore self-sustaining natural populations will be directly tied to how well culture programs maintain the unique balance of polymorphic traits adapting each imperiled population to its natural local environment.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Comparative analysis of skin and gut microbiome dynamics in wild versus farmed Atlantic salmon smolts

Tang, Patrik Anthony[1] (*ptan@norceresearch.no*) Oral Presentation
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Abstract: Atlantic salmon (Salmo salar L.) has a complex life cycle during which smoltification –the morphological, behavioral, and physiological transformation from parr to smolt that prepares the fish to tolerate and thrive in seawater– is a key developmental milestone. In aquaculture, smolt quality is a major issue in production. Smolts experience significant stress when transferred from land-based facilities to sea cages, due to the abrupt change to environments where conditions are less controlled and where they are exposed to an array of new pathogens. Consequently, seawater transfer is a period of heightened economic loss and welfare challenges in the salmon industry. In contrast, wild salmon are generally considered more resilient and adaptable than their farmed counterparts.
Microbial communities occupying mucosal surfaces are central to fish health; they mediate every interaction between the host and environment and contribute to digestion, immunity, and disease resistance. Despite their importance, most studies of salmon microbiota focus on compositional descriptions, with limited insights into host–microbe dynamics.
As part of the HEU2022 project BlueRemediomics, we conducted a proof-of-concept comparative study of skin and gut microbiomes in wild and farmed Atlantic salmon smolts, examining associations with environmental conditions, health, and performance. Our results provide new insights into how domestication and rearing environments influence microbiome assembly, diversity, and functional potential, and identify features of the wild microbiome that may serve as indicators of fish health or resilience. Such insights can contribute to improving the performance and welfare of farmed salmon as an environmental service.
Acknowledgments: Funding: BlueRemediomics-Grant No.101082304, EU Horizon Europe Programme. Farmed fish were sampled from Lerøy´s land and sea facilities in Norway.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Developments in fish swimming technology for high-volume aquaculture systems

Herbert, Neill (*n.herbert@auckland.ac.nz*) Oral Presentation
University of Auckland

Abstract: Research on farmed fish in tanks over the last 50 years tells us that encouraging fish to swim (exercise) fast on a sustained basis often conveys the benefit of improved productivity (better growth and feed conversion efficiency), improved flesh quality and lower levels of stress. But exercise is rarely implemented as an active management strategy in seacage aquaculture because water currents are thought to be the only way of exercising fish yet are impossible to control in these systems. To overcome this limitation, lighting technologies that emit a moving light stimulus are being explored as a means of triggering the innate optomotor reaction of fish, creating controlled schooling/swimming reactions and providing the benefits of exercise under low flow conditions. Video recordings day and night reveal that several farmed fish species (salmon, kingfish and

bream) in tanks and seapens swim and school at speed during the day but they swim slow and non-directionally during dark nighttime periods, whereby the benefits of sustained optimal exercise (improved productivity, quality and welfare) are probably not being gained. Our research demonstrates that fish subjected to moving light stimuli in the dark continue to school in one predominant direction at faster speed and that this swimming behaviour can lead to improvements in growth, FCR and stress reduction. This technology has application for commercial high-volume aquaculture and the implementation of exercise as an active management strategy.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Development of a Gut Dybiosis Assay for Chinook Salmon Oncorhynchus tshawytscha

Vandersteen, Wendy (*wendyv@firstmate.com*) Oral Presentation
Co-Authors: Cannon, Barb; Marty, Gary; Milligan, Barry and Siah, Ahmed
Taplow Ventures Ltd

Abstract: The gut microbiome is an important component of health status through interaction with the immune system, nutrient metabolism, behaviour, and other physiological pathways.Enteritis is a condition impacting the production of salmon.Although the cause of the enteritis is not always known, it is believed that a dysbiosis in the gut microbial communities underlies its development.Aside from impacting aquaculture production through reduced growth and increased mortality, gut enteritis is a fish welfare issue and an environmental issue through reduced absorption of nutrients from food, increased sensitivity to environmental stressors, and increased susceptibility to pathogens. Despite the importance for overall fish health and welfare, there is limited understanding of the functional and taxonomic composition of a healthy microbiome. The overall objective of this project is to provide proof-of-concept of a test to assess gut dysbiosis from a fecal sample.This test will be validated as a screening tool to help fish health professionals make informed decisions on treatment strategies.In addition, the test will facilitate further work to assess the effectiveness of probiotics or other nutritional supplements on maintaining healthy gut microbiomes and allow for further progress towards sustainable use of resources and optimal fish health and welfare.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Joint morphological and DNA metabarcoding gut content analysis reveals adult body size but not biological sex as factors influencing dietary composition divergence in multiple populations of cunner wrasse (Tautogolabrus adspersus).

Bender, Chris (*cbende02@uoguelph.ca*) Oral Presentation
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Abstract: Assessment of a species' potential as a sustainable biological solution for aquaculture pest management depends on the delineation of said species' natural dietary

composition throughout the entirety of its life cycle, given the potentially large dietary shifts that are common amongst many fish species. The cunner (Tautogolabrus adspersus) is a common neritic demersal marine fish being considered as a cleaner fish solution to pest management in Atlantic Canadian aquaculture. Little is known about how its diet changes as a function of size and sex, nor how these factors influence cleaner fish potential on the basis of natural predatory tendencies. This study investigated the effect of size (weight, total length, body condition) and sex on dietary composition of cunner across a 1755-km stretch of the northwestern Atlantic using traditional morphological and dietary DNA metabarcoding methods of gut content analysis. Analysis of diet using ordination (PCA/PCoA) and statistical tests (PERMANOVA) revealed that size metrics like weight and length significantly influence the diet of this species. Small cunner contained more soft-bodied organisms (ascidians, anemones), whereas larger cunner guts contained more hard-shelled organisms (bivalves, crustaceans). These results highlight significant changes in prey consumption as a result of size in this species, demonstrating how secondary consumers can vary in how they link different parts of marine food webs within a single individual's lifetime. Results also highlight how, in the context of cleaner fish applications, a mixture of sizes of adult cunner may be more successful at reducing pest abundances, due to shifts in dietary composition as a result of size.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Farmed sea lettuce (Ulva fenestrata) as a promising ingredient for rainbow trout (Oncorhynchus mykiss)

Andersson, Marica[1] (*marica.andersson@bioenv.gu.se*) Oral Presentation
Co-Authors: Warwas, Niklas [2]; Hinchcliffe, James[3]; Johansson, Elin[1]; Roques, Jonathan A. C.[1]; Gunnarsson, Pontus[4]; Stedt, Kristoffer[1]; Steinhagen, Sophie[1]; Pavia, Henrik[1] and Sundell, Kristina[1]
[1] University of Gothenburg; [2] Skretting, Stavanger; [3]Marine Ingredients Denmark, [4]Swedish University of Agricultural Sciences

Abstract: Introducing novel feed ingredients is essential to ensure a sustainable expansion of the aquaculture industry. Seaweeds, such as the green alga sea lettuce (Ulva fenestrata), are promising ingredients as they absorb nutrients directly from the seawater and thereby have a low environmental impact. They also have a well-balanced amino acid profile and high content of bioactive compounds and micronutrients. Despite these promising traits, the effects of farmed sea lettuce in feed for cold-water salmonids has not yet been studied. Here we evaluated how the health and welfare of rainbow trout (Oncorhynchus mykiss) was affected by including Ulva meal in their feed. The study lasted for 12 weeks, and experimental diets included a control feed, and two feeds with 3% and 15% Ulva meal, respectively. We found the highest growth rate in fish given the 15% Ulva diet during the first six study weeks and in the 3% Ulva diet during the last six study weeks. This was likely due to an increased feed intake and improved palatability from the Ulva inclusion. Analysis of intestinal health parameters, primary barrier function and histomorphology, revealed no impaired barrier or inflammatory responses to any of the

diets. Similarly, the stress indicators, including plasma cortisol levels and oxidative stress markers in liver and muscle, remained unaffected across diets. Interestingly, the fillet colour became more yellow with increased Ulva inclusion, which could potentially negatively influence consumer acceptance. Nevertheless, the results demonstrate that sea-farmed Ulva can contribute to improving the sustainability of future aquafeeds for rainbow trout.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Dietary and environmental interventions to optimize growth, stress, and reproductive outcomes in cultured longfin yellowtail, Seriola rivoliana

Chang, Ryan (*chang25@hawaii.edu*) Oral Presentation
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Abstract: The longfin yellowtail (Seriola rivoliana) is an emerging candidate for mariculture around the world; however, many producers rely on wild caught fish. Captive breeding and production of these fish leverage both diet and environmental temperature, among other factors, to modulate growth, energy utilization, and reproductive performance. However, since the optimal rearing practices for yellowtail broodstock are unresolved, we conducted a longitudinal trial in which we raised juvenile yellowtail to sexual maturity on two diets for two years. Fish were fed either a commercial pellet-based diet or a freshseafood-based diet, consisting of gelatinized mixture of fish and squid with nutrient supplementation (gel) for 5 months, followed by a whole fresh seafood diet (whole). We then exposed sexually mature fish from each dietary regime to a transient reduction in temperature to probe for signs of reproductive development. We found that fish fed the gel diet grew slower than those fed pellets; growth increased when fish switched from gel to a whole diet, equaling and surpassing that of the pellet diet. Fish fed gel had lower hepatosomatic index and were more susceptible to repeated handling stress in a 24 h experiment. In reproductively mature fish, cold exposure increased gonadosomatic index in males. The current data supports the utility of dietary and temperature interventions to modify growth, stress, and reproduction. Furtheridentification of other phsyiological traits and pathways will ultimately provide insight into how diet and temperature modulate the health and production of captive yellowtail and lay the foundation for developing selective breeding strategies.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Fasting reduces intestinal tissue mass and transport capacity in salmonids

de Fonseka, Raneesha[1] (*raneesha.de.fonseka@bioenv.gu.se*) Oral Presentation
Co-Authors: Sundell, Kristina[1]; Jönsson, Elisabeth[1]; Nilsen, Tom Ole[2]; Takvam, Marius[2]; Vikesåe, Vibeke[3] and Sundh, Henrik[1]
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Abstract: Short-term fasting is routinely applied in aquaculture prior to many handling events. However, the effects of fasting on intestinal transport capacity remain poorly understood. In two experiments, we investigated the effects of 4-5 days of fasting on intestinal mucosal mass and transport capacity in Atlantic salmon smolts and in freshwater rainbow trout using the gut-sac method. In Atlantic salmon smolts, intestinal mass and transport were assessed after 4 days of fasting before seawater transfer. In rainbow trout, the same parameters were assessed daily during 5 days of fasting. Atlantic salmon smolts lost >12% of mucosal weight and showed a >19% reduction in fluid transport capacity, which persisted for at least up to 6 weeks in seawater. Smolts fed a high-lipid diet (5% extra fat than the control) had recovered after these 6 weeks of re-feeding. In rainbow trout, mucosal tissue had regressed from day 3 onwards (>14%), along with a reduction in transport capacity. The rainbow trout intestine recovered after 5 days of re-feeding. These findings demonstrate that short-term fasting compromises intestinal mucosal mass and transport capacity, thereby negatively affecting nutrient uptake and osmoregulation, particularly during sensitive life-stage transitions like smoltification. A high-lipid diet during smoltification can protect the fish from the negative effects of fasting. In rainbow trout, it seems safe to fast the fish for up to 2 days with regard to intestinal transport capacity. Our results highlight the importance of physiological studies to evaluate common aquaculture practices to improve farming strategies without compromising health and welfare.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Use of alternative insect feed ingredients in sustainable wolf eel aquaculture

Dykstra, Jeremy[1] (*jdykst01@student.ubc.ca*) Oral Presentation
Co-Author: Niemann, Benjamin[1]; Vadersteen, Wendy[2]; Forster, Ian[3] and Frommel, Andrea[1]
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Abstract: Aquaculture is a rapidly growingprotein-producing agriculturalsector;however,the use ofalternativeingredients in formulated fish feedsshouldbe investigated toimprovesustainability.Insectmeal is a novel animal feedingredientthat offers a carbon-neutral, healthy protein alternative to fishmeal and plant-based products. Insects, such as black soldier fly larvae (BSFL,Hermetiaillucens), can be farmed on consumer food waste, contributing to a circular economy. The wolf eel (Anarrhichthysocellatus), a fish native to the Pacific Northwest, shows strong potential forhigh inclusion of BSFL

initsdiet,asitprimarilyeatschitin-containinginvertebrates in the wild.Fourisoproteinous,isolipidic,and isocaloric diets,including a control, 10%, 20%, and 40% protein replaced with defatted BSFL,weretestedduringa 3-monthperiodon juvenile wolf eel.Fish growth,condition factor, proximate composition(protein, lipid, fatty acids), chitinase activity, and histopathologyare beingassessed.We expect the 40% BSFLdiettoyieldthe highest specific growth rates,condition factor,and chitinase activity, whilepossessinga balanced nutritional profile with minimal histopathological concerns.This research advances our physiological understanding of wolf eelandcontributes tosustainable aquaculturediversificationthrough thisemergingalternativemarinespecies.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Enhancing growth, coloration, and fitness of koi (Cyprinus rubrofuscus) with purple bacteria

Opinion, April Grace (*aprilgrace.opinion@uantwerpen.be*) Oral Presentation
Co-Authors: Diaz Allegue, Luis; De Bie, Maxim; Vlaeminck, Siegfried; De Boeck, Gudrun
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Abstract: Ornamental aquaculture is a rapidly expanding sector that can reach exceptionally high economic value, making nutrition-driven improvements in growth, appearance, immunity, and fitness commercially relevant. Conventional pigment enhancers and fishmeal-dependent diets raise sustainability and supply volatility concerns, prompting interest in alternative microbial-based ingredients. Purple bacteria (PB), in this case Rhodobacter capsulatus, represent promising multifunctional feed ingredients, providing high-quality protein alongside carotenoids and other antioxidative compounds. Previous work demonstrated consistent positive effects, including enhanced growth, pigmentation, and disease resistance in guppy and shrimp fed with PB-based diets.

The present study investigated PB supplementation in koi, Cyprinus rubrofuscus, an economically important ornamental species. Fish were randomly assigned to five dietary treatments: a negative control without additives; a positive control containing a pigment-rich blend of Chlorella and Moringa; and experimental diets including 2%, 5%, and 10% PB. After a 20-week feeding experiment, growth, coloration, aerobic performance, and swimming capacity were assessed and compared among treatments. Complementary analyses, including pathogen-challenge tests, plasma metabolomics, and gut microbiome analyses, are ongoing to evaluate immune resilience and elucidate mechanisms.

Results showed that PB-fed koi exhibited superior growth, with the 2% inclusion level producing the highest growth rate and condition factor. PB supplementation also altered skin hue and lightness, resulting in a highly valued brighter appearance. Respirometry trials further revealed enhanced aerobic scope and swimming performance, reflecting improved fitness. These findings position PB as a sustainable, multifunctional feed ingredient that improves growth, coloration, and physiological performance while reducing dependence on fishmeal and synthetic or plant-based pigments.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Insect-based diets modulate welfare, health, and gut microbiome functions in farmed atlantic salmon

Tang, Patrik Anthony[1] (*ptan@norceresearch.no*) Oral Presentation
Co-Authors: Gilannejad, Neda[1]; Menanteau-Ledouble, Simon[1]; Gharbi, Naouel[1]; Rostad, Kristine[1]; Bø-Randulff, Marie[1]; Røise, Dagne[1]; Nilsen, Marius[1]; Tronci, Valentina[1]; Gorissen, Marnix[2]; Ebbesson, Lars[1]; Ghemis Malcic, Radu[3]; Sánchez Cueto, Pablo[3]
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Abstract: Continued expansion of aquaculture requires diversification of feed ingredients to ensure continued supply of sustainable ingredients. Insects are highly desirable based on their high protein content and suitable amino acid composition and can be produced using side-stream agro-industrial wastes. The diversity of insect species provides an array of nutrient profiles that can be optimized into blends of species to match the requirements of farmed fish. While studies confirm replacement using a single insect species can fulfill requirements to maintain growth, and digestibility, less attention has been given to other important physiological parameters necessary for a good health, welfare, and resilience during the challenging conditions such as the freshwater (FW) to seawater (SW) transition phase in Atlantic salmon (Salmo salar). Here, we evaluated how salmon stress responses and osmoregulatory capacities are affected by substituting soy protein concentrate (SPC) with different inclusions (3%, 25%, and 50%) of a novel mix of insect meal (IM) [black soldier fly larvae (Hermetia illucens) and yellow mealworm (Tenebrio molitor)]. The feed trial included 8 weeks in FW and transfer to SW for 6 weeks. At the end of both FW and SW phases, fish were subjected to an acute challenge test (ACT), by high density, to monitor stress responses capacities. The gut microbiome metatranscriptome was analyzed and correlated with fish health and welfare indicators. Plasma levels of cortisol, ions, and other energy status and stress metabolites were quantified, complemented using expression levels of genes involved in gill osmoregulation, brain plasticity, and systemic and local immunity. Our study provides a novel and holistic framework for evaluating the dietary impacts of novel feed ingredients on health, welfare, and microbiome function in salmon.
Acknowledgments: Funded by EEA InFishMix-PT-INNOVATION-0094 and HEU2022 BlueRemediomics-Grant No.101082304.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Effects of exercise training on cardiac morphology of rainbow trout

Anttila, Katja[1] (*katant@utu.fi*) Oral Presentation
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Abstract: Farmed fish are suffering from significant cardiac malformations which is a serious welfare issue in aquaculture. The altered morphology of heart is connected to reduced swimming capacity and also to deaths related to stress and diseases. The atherosclerosis is, furthermore, a substantial problem in aquaculture and significant number of farmed fish are having coronary lesions which negatively affects the blood delivery to cardiac tissues. The lockage of blood delivery to heart might even imperil the fish e.g. to heat waves. Thus, besides welfare of fish, the cardiac malformations might affect the economy of fish farming.
Fortunately, the heart of the fish is extremely plastic and even morphology can change via environmental signals. In this project our goal was to investigate if increasing the swimming activity in aquaculture tanks via exercise training could influence the morphology of heart of farmed rainbow trout and make it resembling more the ones of wild fish. The fish were trained for 6 hours per day with velocity of 0.9 body lengths per seconds (bl/s) while control fish swam 0.3 bl/s. For trained fish the velocity was reduced to control level after the training session. The training program lasted 4 weeks after which the cardiac morphology was investigated and severity of atherosclerosis was analyzed.
The results of the project are not important only scientifically but they are important for aquaculture as well since we should try to seek different ways to enhance the welfare and health of aquaculture fish.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Osmorespiratory compromise in triploid (sterile) salmonids

Benfey, Tillmann[1] (*benfey@unb.ca*) Oral Presentation
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Abstract: Triploidy induction is the only practical method for producing reproductively sterile populations of fish for aquaculture and fisheries management, but triploids are rarely used for this purpose because they tend to perform poorly under stress, for instance when reared at elevated temperature or low oxygen. This is consistent with evidence that triploids have reduced aerobic scope. As part of a larger research program investigating environmental tolerances of triploids, we have been examining their ability to cope with the competing requirements of meeting oxygen demand while minimizing rates of ion diffusion across the gill lamellae, i.e., the osmorespiratory compromise. This presentation will present results from two such studies. In the first, we show that interlamellar cell mass

(ILCM) size is significantly larger in triploid brook charr, Salvelinus fontinalis, than sibling diploids under both normoxia and moderate hypoxia, implying that triploids have reduced functional lamellar surface area for oxygen uptake. This was realized by reduced acute hypoxia tolerance of these same triploids in comparison to the diploids. In the second study, we show that triploid rainbow trout, Oncorhynchus mykiss, have a larger ILCM and higher plasma osmolality and ion levels (Na+, Cl-, K+), on average, than sibling diploids when reared at high temperature in full-strength seawater, although none of these differences were statistically significant. However, taken together, they suggest that triploids need to invest more than diploids into protecting themselves from osmotic distress and thereby compromise their ability to obtain sufficient oxygen to meet metabolic demands at high temperature.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Temperature and ploidy effect on red blood cell physiology in brook charr (Salvelinus fontinalis)

Virag, Victoria (*victoria.virag@unb.ca*) Oral Presentation
Co-Author: Benfey, Tillmann
University of New Brunswick

Abstract: Triploidy, the presence of three sets of chromosomes, is easily induced in fish and results in sterility. This is valuable for aquaculture as it prevents reproduction and improves genetic containment. However, triploid fish often perform poorly in warm, hypoxic conditions which may be due to their lower erythrocyte surface area to volume ratio (an outcome of their larger cell size). A common cellular response to temperature change is to alter membrane lipid composition to maintain appropriate membrane fluidity (i.e., homeoviscous adaption), disturbing cell shape and trans-membranal molecular transport systems. The objective of this study is to test the hypothesis that the increased erythrocyte size in triploids compromises the ability of the cells to withstand osmotic challenges and to transport respiratory gas efficiently due to altered cell membrane lipid composition at a given temperature and a reduced ability for homeoviscous adaption. To test this, sibling diploid (control) and triploid brook charr (Salvelinus fontinalis) will be acclimated to 12℃, 15℃, and 18℃, with 15 ℃ being the diploid optimum. Blood samples will be collected to compare (1) membrane lipid profiles using gas chromatography to examine fatty acid composition, (2) ability of erythrocytes to maintain cellular integrity during osmotic stress (by determining the osmotic pressure at which the erythrocyte rupture, i.e., hemolysis), and (3) rate of erythrocyte oxygen uptake (in vitro) using a temperature-controlled 24-well microplate respirometry system. Data collection is currently in progress.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Effect of environmental variation on the regulation of skin barrier function in rainbow trout in aquaculture

von Ehr, Michelle (*michelle.vonehr@gmail.com*) Oral Presentation
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Abstract: Global aquaculture production is constantly increasing, and so is the need for sustainable practices that ensure fish health and welfare. The skin is a primary epithelial barrier and therefore a critical interface between the organism and its surrounding environment. Tight junctions maintain epithelial integrity by alternating their trans-membrane proteins, which exist in barrier-forming and pore-forming isoforms. This regulates the skin's paracellular permeability and selectivity. In monolayered epithelia such as the gills, tight junction morphology differs with salinity, appearing "deep" in freshwater-acclimated fish and "shallow" in seawater-acclimated fish, corresponding to high and low barrier functions, respectively. However, the localization and regulation of tight junctions within stratified epithelia such as the skin remain poorly understood. This project investigates skin barrier function in rainbow trout, focusing on tight junction localization, morphology, and regulation in response to variable salinity. Barrier properties are assessed ex vivo using Ussing chamber methodology to measure transepithelial electrical resistance under different salinities and ion-substitution conditions. Tight junction localization and morphological changes are examined by transmission electron microscopy. Preliminary results suggest that environmental salinity above 2 ppt reduces skin resistance, indicating a possible shift toward shallower, more permeable tight junction complexes. These findings provide early insight into how external environmental factors impact skin barrier integrity in salmonids. An improved understanding of tight junction organization and regulation in fish skin is essential for elucidating how common husbandry conditions influence barrier function and resilience, thereby promoting fish welfare in aquaculture.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Nephrocalcinosis in a jar: a multi-factor in vitro model of CO_2 induced nephrocalcinosis

Emadi, Cameron[1] (*cameron.emadi@ubc.ca*) Oral Presentation
Co-Authors: Wood, Chris[1]; Vik, Jon Olav[2]; Takvam, Marius[3]; Nilsen, Tom Ole[3] and Brauner, Colin J.[1]
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Abstract: Nephrocalcinosis ("kidney stones") is a persistent health challenge in intensive aquaculture systems, affecting fish welfare and productivity. Although its causes remain poorly understood, it is most prevalent in recirculating aquaculture systems (RAS), where fish experience chronically elevated dissolved CO2 and O2. These conditions induce

sustained hypercapnia and hyperoxia that disrupt acid-base regulation. While nephrocalcinosis is associated with RAS environments, the mechanism linking environmental gas fluctuations to renal precipitation remains unresolved. To isolate physiological drivers of nephrocalcinosis, we developed an in vitro model using synthetic pre-urine formulated to match fish pre-urine ionic composition. The pre-urine was sequentially subjected to: (1) elevated PCO2 (1–3%) to mimic hypercapnia-induced respiratory acidosis; (2) bicarbonate addition to simulate metabolic compensation; and (3) rapid PCO2 reduction reflecting routine aquaculture events (e.g., transfer or handling). Precipitation, quantified by turbidity, was rapid and progressive following the abrupt PCO2 decrease (step 3 above). The magnitude of alkalinization (ΔpH) strongly predicted precipitation intensity, especially at higher temperatures (18°C > 12°C > 6°C). Ionic manipulations of pre-urine further demonstrated that Ca2+, Mg2+, and PO43- concentrations shift precipitation thresholds under identical CO2 treatments, indicating that renal mineralization risk emerges from the interaction between acid-base status, temperature, and ionic loading. These findings support a mechanistic framework in which fluctuations in environmental CO2 drive renal alkalinization events that promote mineral precipitation in urine, identifying acid-base instability, not simply elevated CO2, as a potential trigger of nephrocalcinosis and underscoring the need to integrate environmental physiology with renal chemistry in intensive aquaculture systems (FHF-Norway).

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Set in stone: The effect of diet and carbon dioxide on nephrocalcinosis development

Mullins, Julia[1] (*julia.mullins@skretting.com*) Oral Presentation
Co-Author: Le, Thi Hong Gam [1]; Thomsen, Taarn[2]; Yousaf, Muhammad Naveed[1]; Nilsen, Tom Ole[2]
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Abstract: Nephrocalcinosis, commonly known as kidney stones, is a key production challenge facing the salmonid industry. The condition is characterised by the deposition of calcium and magnesium mineral salts in the kidney, leading to tubule dilation, tissue degeneration, and loss of renal structure. Nephrocalcinosis has been observed in fish ranging from 2 g in freshwater, to 6 kg in saltwater, and occurs in both flow-through and recirculating aquaculture systems (RAS). It is associated with increased mortality, compromised welfare, reduced growth, and poor performance after transfer to seawater. The main aim of this experiment was to evaluate the impact of diet and carbon dioxide (CO_2) on the development of nephrocalcinosis in Atlantic salmon reared in freshwater RAS. Fish were distributed among 12 RAS tanks. Six tanks were exposed to three alternating periods of elevated CO_2 (30 mg/L) followed by periods of low CO_2, while six tanks were maintained at CO_2 levels below 5 mg/L. Within each CO_2 regime, fish were fed one of three diets containing normal, medium, or high levels of mineral salts, with treatments replicated in duplicate.
Physiological and anatomical parameters, including gross and microscopic kidney scores, growth performance, and blood and urine parameters were assessed across acute

exposure and recovery time points. The combination of elevated CO_2 and high dietary mineral levels resulted in kidney damage and alterations in several blood and urine parameters. These findings highlight the importance of integrated RAS management linking water chemistry and dietary mineral control to reduce nephrocalcinosis risk and improve fish performance.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Microbiome Effects of Fulvic and Humic Acid Diets and Baths

Niemann, Benjamin[1] (*benjien@student.ubc.ca*) Oral Presentation
Co-Author: Vandersteen, Wendy[2]; Frommel, Andrea[1]; Forster, Ian[3]; Guan, Leluo[1]; Shimomura, Miki[3] and Parfrey, Laura Wegener[1]
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Abstract: Aquaculture is a fast-growing protein production sector in agriculture, but there are major sustainability concerns in terms of disease management within the context of the wider environment. Commonly, antibiotics are added directly to feed which can then decay in the natural ecosystem, fostering resistance in microbial populations; immunostimulants and other 'functional feeds' offer an alternative to this. Fulvic and humic acids, which are classifications of humic substances (HS), are a type of immunostimulant isolated from natural organic carbon in soils. HS addition is being pursued in both field crop and animal husbandry agriculture to increase productivity, so the goal of these experiments was to evaluate if HS use could be of benefit to local Canadian aquaculture operations. Over two years, we ran eight experiments to compare the effects of HS additives in two forms (diet additive and liquid) in aquaculture for four fish species farmed here in Western Canada (Oncorhynchus mykiss and tschawytscha, Salvelinus alpinus, and Anoplopoma fimbria). We sampled the gut contents to examine effects of the diet and skin mucosa samples to determine effects of the liquid supplement, in addition to a variety of physiological parameters to evaluate effects on the host. Overall, there was no effect of HS treatment on either host physiology or community diversity and composition. The largest effect on community composition was attributed to species differences.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Interactive effects of warming and dietary nanoplastics on erythro-immune homeostasis in gilthead seabream (Sparus aurata)

Mancera Romero, Juan Miguel [1] (*juanmiguel.mancera@uca.es*) Poster Presentation
Co-Author: Carrillo, Mercedes [1]; Hachero, Ismael [2];
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Abstract: Ocean warming and nanoplastic (NP) contamination represent converging stressors for marine aquaculture. In eurythermal species such as gilthead seabream

(Sparus aurata), elevated temperature increases metabolic demand and oxygen consumption, potentially reshaping erythro-immune homeostasis. However, the combined effects of warming and chronic dietary NP exposure on physiological status of S. aurata remain poorly resolved. Juveniles were reared for 49 days in a recirculating aquaculture system (initial density: 3.57 kg m^{-3}) under a 2×2 factorial design including two temperatures (18 and 26 °C) and two diets (control and NP-supplemented: 50 mg NP [50 nm, reference PS02002, Bangs Labs]/kg aquafeed). Temperature emerged as the dominant driver of erythrophysiological modulation. Fish maintained at 26 °C enhanced red blood cell count (RBC), hemoglobin (HGB), and hematocrit (HCT) ($p < 0.0001$), consistent with enhanced oxygen-carrying capacity under increased metabolic demand. MCV and MCH were also temperature-dependent ($p < 0.05$), whereas MCHC decreased at elevated temperature ($p < 0.0001$), indicating adjustments in erythrocyte dynamics rather than anemia. Red cell distribution width (RDW-CV) was affected by temperature, diet, and their interaction ($p < 0.0001$), suggesting subtle alterations in erythropoietic regulation under combined stressors. Dietary NP exposure significantly increased non-erythrocytic and mononuclear cell counts ($p < 0.0001$), while granulocyte proportion was influenced by diet and temperature–diet interaction ($p < 0.05$), evidencing immunomodulatory effects. Overall, warming primarily remodel erythrophysiological parameters, whereas dietary NP exposure modulated immune-related hematological indices. These findings reveal differential yet potentially complementary impacts of climate-driven warming and NP contamination on systemic homeostasis in marine aquaculture species.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Alternative vectors for fish nutrition: “mush” room for improvement?

Seethram, Maya (*mayaseethram@icloud.com*) Poster Presentation
Co-Author: Zink, Lauren; Lin, Nicholas; Wood, Chris and Hallam, Steven
The University of British Columbia

Abstract: In high-density fish aquaculture settings, maintaining water quality while feeding to promote growth is a major challenge; uneaten food often results in spikes of ammonia and nitrites. Therefore, development of alternative aquaculture feeds presents opportunities to promote sustainability, reduce operational costs, and optimize fish nutrition. Fungal mycelium represents an easily digestible protein source that can be produced at high volumes without disproportionate negative impact to the environment.Despite its ease of production and compelling nutritional profile, mycelium has yet to be applied as an effective nutrient vector in aquaculture.Our team has created a novel mycelium-based (Pleurotusostreatus) diet which does not negatively impact water quality. In preliminary trials, we incorporated nutrient-dense inclusions to our mycelium vector. The resultant feeds exhibit several desirable characteristics: (1) neutral buoyancy to permit longer feeding periods for pelagic species, (2) high structural integrity allowing the feed to remain intact to optimize grazing; and (3) a high degree of customizability to manufacture different feed sizes and shapes for various species.In the present study, we discuss how the feeding of mycelium, and mycelium with additions of cyanobacteria

(Sodalinema yuhuli AB48) or brine shrimp (Artemiasalina) affect both juvenile and adult zebrafish (Danio rerio). We assessed body composition, growth rate, hypoxia tolerance (as a physiological metric), and activity (as a behavioural metric) to evaluate the role of a mycelium-based food delivery system in preserving water quality while meeting the nutritional requirements of fish relevant to sustainable aquaculture practice (UBC MCELLS Network, NSERC).

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Biological authentication and traceability tools to strengthen seafood supply chain transparency: insights from the FishEUTrust project

Tang, Patrik Anthony[1] (*ptan@norceresearch.no*) Poster Presentation
Co-Author: Gharbi, Naouel[1]; Menanteau-Ledouble, Simon[1]; Ogrinc, Nives[2]
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Abstract: Seafood mislabelling and limited traceability remain significant challenges for global seafood markets, affecting consumer confidence, fisheries management, and sustainability efforts. Biological tools for species authentication and product verification are increasingly important to ensure the integrity of seafood supply chains. The EU-funded FishEUTrust project addresses these challenges by integrating biological sampling, molecular identification methods, and traceability approaches to strengthen trust in seafood products.

Within FishEUTrust, biological samples from multiple seafood value chains, including aquaculture species such as Atlantic salmon, are collected to support the development and validation of authentication tools. Molecular approaches, including DNA-based species identification and biomarker analyses, are applied to verify species identity, detect potential mislabelling, and support traceability efforts. These biological methods provide robust reference data that can be linked to digital traceability systems and supply-chain information.

The project also investigates how biological verification methods can be integrated into broader traceability frameworks to improve transparency and accountability in seafood markets. By combining biological authentication tools with traceability technologies, FishEUTrust aims to strengthen the scientific basis for seafood verification and support more reliable product information.

This work highlights the important role of fish biology in supporting trustworthy seafood systems and demonstrates how biological data can contribute to improving traceability, reducing fraud risks, and reinforcing consumer confidence in seafood products.

Acknowledgment: This work was supported by the European Union's Horizon 2020 research and innovation programme through the FishEUTrust project.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Physiological mechanisms of reduced feed intake and growth after vaccination in salmonid fish

Wallbom, Nicklas[1] (*nicklas.wallbom@bioenv.gu.se*) Poster Presentation
Co-Author: Jönsson, Elisabeth[1]; Sandblom, Erik[1]; Sundh, Henrik[1] and Khansari, Ali[2]
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Abstract: Immune activation following infection and vaccination can reduce feed intake and growth in salmonid fish, but the physiological mechanisms are poorly understood. Our recent research has consistently shown that vaccination neither increases basal metabolic costs nor impairs respiratory performance in rainbow trout (Oncorhynchus mykiss) reared in fresh- and sea water. We therefore hypothesized that reduced growth is a mechanism to buffer the available aerobic energy for immune function even when constitutive costs are low. Consistent with that hypothesis, the present study demonstrates that growth rates decrease in a dose-dependent manner in vaccinated trout. Reducing costs associated with feeding, digestion and foraging are other viable short-term strategies to buffer energy availability during immunological challenges. Indeed, we observed temporary reduction in appetite after vaccination. We are currently analyzing expression patterns of central and peripheral hormones and neuropeptides to test whether vaccination affects appetite regulation. In addition, we are analyzing the expression patterns of pro-inflammatory cytokines to assess potential immune-endocrine pathways that mediate such responses.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Cardiac morphology as an indicator for growth in farmed Atlantic Salmon (Salmo salar)

Wong-Branscome, Olivia (*olivia.wong@maine.edu*) Poster Presentation
Co-Author: Hamlin, Heather
University of Maine

Abstract: Premature sexual maturation found in farmed Atlantic salmon (Salmo salar) is a significant challenge for the rising aquaculture industry as it reduces somatic growth, harvest yields, and production efficiency. While cardiac morphology in aerobic performance, metabolic allocation, and life history is documented across fish species, the relationship between heart structure and growth and/or maturation within a salmon aquaculture context remains under-studied. This study aims to investigate whether gross cardiac morphology and ventricular tissue composition can serve as an indicator for growth and reproductive investment in farmed Atlantic salmon.
With the use of a dataset of several hundred intact and fixed hearts collected from standardized USDA-reared fish, we aim to quantify cardiac variation through multi-angle digital photography and morphometric analysis in ImageJ. From this, we will extract ventricular dimensions, bulbus arteriosus measurements, and tissue compositions. These data will be integrated with extensive individual metadata, including body weight, total length, gonadosomatic index, sexual maturity stage, sex, and family line.

We hypothesize that variation in gross heart morphology indicates an energetic trade-off between somatic growth and reproductive development and that enlarged hearts will show unique pathological signs. Data collection and morphometric analysis is currently ongoing and primary findings have not yet been established. Final results from this work are expected to show the first integrative characterization of cardiac morphology in relation to maturation and energy allocation in aquaculture-reared Atlantic salmon with implications for selective breeding, stock management, and animal welfare monitoring.

AQUACULTURE Applying knowledge of fish biology to sustainable aquaculture

Can exercise training improve mitochondrial function in rainbow trout cardiomyocytes?

Anttila, Katja[1] (*katant@utu.fi*) Poster Presentation
Co-Authors: Henttinen, Tiina[1]; Le, Thuy[1]; Seppänen, Eila[2]; Ekström, Andreas[3] and Haverinen, Jaakko[4]
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Mortalities in finfish aquaculture are increasingly reported during summer months due to climate change. There is a need for adaptive strategies to enhance survival of farmed fish under heat stress. As the heart is one of the first organs failing during heat stress, it presents a target for improving thermal tolerance. This is particularly relevant for farmed fish given their baseline cardiovascular performance is already compromised compared to wild conspecifics, potentially predisposing them to heat-wave mortality. Cardiovascular performance during heat stress can, however, be enhanced through exercise training, and our team has recently demonstrated this in farmed rainbow trout.
Building on this, our project aims to uncover the physiological mechanisms underpinning exercise-enhanced thermal tolerance, beginning with mitochondrial performance. We hypothesised that increased mitochondrial function efficiency may contribute to improved cardiac performance under heat stress. To test this, we reared two groups of farmed rainbow trout under standard aquaculture conditions: a control group exposed to standard water flow (0.5 body lengths per second; bl/s), and an exercise-trained group exposed to increased flow (0.5–1.3 bl/s) for six hours daily over six weeks. We measured mitochondrial ATP production efficiency in cardiac tissue at optimal (17 °C) and cardiac arrhythmia-inducing (23 °C) temperatures.
Our results demonstrate that exercise training made slight improvements in mitochondrial function efficiency at critical temperatures, which could lend support to improved cardiac function under heat stress. For finfish farmers, exercise-training presents as a promising, non-invasive tool to improve production stock stressor tolerance and welfare.

WELFARE Using behaviour and physiology to understand fish welfare

Using behaviour and physiology to understand fish welfare

Katherine Sloman and Lynne Sneddon

Abstract: The way that humans interact with fishes, and what fishes may experience through those interactions, often raises scientific, political and public concern in relation to their welfare. Assessment of fish welfare often involves observation of behavioural changes or deficits, and draws on our understanding of the way that fishes respond physiologically to environmental challenge. In reality, combined measures that integrate behaviour and physiology are likely needed to improve welfare assessment. Fishes represent important scientific models for human health, a key source of protein for human consumption through aquaculture and fisheries and a popular focus of human enjoyment through sport fisheries, home and public aquaria. This session aims to discuss how we can better understand fish welfare across this range of scenarios, but also to share welfare refinements from different fields.

WELFARE Using behaviour and physiology to understand fish welfare

Melanin-based skin pigmentation as an indicator of stress and welfare in Atlantic salmon (Salmo salar)

Gunnes, Mikkel[1] (*mikkel.gunnes@nmbu.no*) Oral Presentation
Co-Authors: Laique, Talha[1]; Gundersen, Hannah N.[1]; Folkedal, Ole[2]; Nilsson, Jonatan[2]; Sørensen, Christina[3] and Øverli, Øyvind[1]
[1]Norwegian University of Life Sciences, [2]Institute of Marine Research, [3]Norwegian Institute for Water Research

Abstract: Assessing welfare in intensive salmon aquaculture requires indicators that reflect underlying neurobiological state in addition to external injury. In this context, melanin-based skin pigmentation in salmonids has been linked to stress responsiveness and coping ability, yet the neurochemical basis of dynamic pigmentation change remains unresolved.We tested whether stress-induced changes in melanin-based skin spots reflect regional serotonergic activity in Atlantic salmon. PIT-tagged fish were photographed before and after overnight confinement, and opercular spot intensity was quantified using automated computer vision–based grayscale analysis. Serotonergic markers (5-HT, 5-HIAA) were measured in brain stem and telencephalon using HPLC.Confinement induced significant within-individual fading of melanin-based skin spots, and stressed fish had paler spots than controls. Spot pigmentation was associated with serotonergic tonein both brain regions, but in opposing directions. Higher brain stem 5-HT was associated with lighter spots at the time of sampling. In contrast, higher telencephalic 5-HT was associated with darker skin spots at the time of sampling, and smaller within-individual spot fading in response to stress. Higher condition factor was similarly linked to darker skin spots.These

results indicate that skin spot dynamics integrate region-specific serotonergic activation and physiological condition rather than reflecting stress exposure alone. Together, these findings provide neurobiological grounding for melanin-based skin pigmentation and extend previous work linking melanin expression to stress-related traits in salmonids.

WELFARE Using behaviour and physiology to understand fish welfare

A fluttering heart: Use of biologgers during behavioural stress tests in rainbow trout

Sneddon, Lynne (*lynne.sneddon@bioenv.gu.se*) Oral Presentation
Co-Author: Kasiouras, Eleftherios
University of Gothenburg

Abstract: Many fish species exhibit divergent phenotypes, ranging from bold to shy, which correlate with cortisol production during the stress response. To determine whether boldness affects the sympathetic nervous system (SNS), heart rate was measured as a proxy for SNS responses during three potentially stressful events. Firstly, rainbow trout (Oncorhynchus mykiss) were classified as bold or shy, using a novel object test. Subsequently, trout were implanted with biologgers to record heart rate in vivo at rest, during and after the behavioral tests. Following recovery, the fish underwent a second novel object test, a confinement test, a pair-wise contest, and a final novel object test to explore the degree of boldness over the experimental period, which remained consistent. Heart rate was relatively higher in both bold and shy animals during the confinement test and the pair-wise contest compared with the novel object test, which indicated that heart rate monitoring was a valid gauge of the valence of the experience. Heart rate responses did not differ between bold and shy trout, indicating that behavioral phenotype did not influence the autonomic stress response. Monitoring stress in captive fish is crucial for their welfare. Our study demonstrates heart rate is a reliable indicator of stress without the need to account for intra-specific bold-shy variation.

WELFARE Using behaviour and physiology to understand fish welfare

From signals to states: biologger-based classification of seabass welfare states in sea-cages

Hoyo-Alvarez [1], Esther (*ehoyo@imedea.uib-csic.es*) Oral Presentation
Co-Author: Cabrera-Álvarez, Maria J. [2]; Vanrell-Valls, Margalida[3]; Palstra, Arjan P.[4] and Arechavala-Lopez, Pablo[1]
[1] IMEDEA (UIB-CSIC); [2] Fish Etho Group Association, [3]LIMIA-IRFAP, [4]Wageningen University & Research

Abstract: Rapid expansion of aquaculture has increased the need for advanced monitoring techniques to ensure fish welfare and optimise management practices. Understanding fish responses to environmental and anthropogenic factors is key to improving welfare standards. Biologgers measuring heart rate (HR) and external acceleration (ACC) offer valuable insight into physiological and behavioural dynamics.

In this study, HR and ACC were recorded from adult European seabass implanted with biologgers and monitored in sea-cages for two 14-day periods in March and July. Feeding and cage maintenance occurred Monday to Friday, while weekends involved no aquaculture-related human activity. A Random Forest (RF) model, trained with labelled data from controlled stress-challenge experiments, classified four welfare states: resting, regular activity, reactive response, and proactive response. Standardized ACC was the main predictor of proactive responses, whereas standardized HR contributed most to resting and reactive states.
Applying the model to sea-cage data revealed clear diel patterns: resting and regular activity predominated at night and early morning, while proactive responses increased from midday, linked to feeding routines. Significant differences emerged between weekdays and weekends, with stress-related states more frequent during weekdays and resting and regular activity dominating weekends, reflecting routine operations and human presence. Seasonal patterns showed higher HR and more proactive responses in July, likely driven by elevated water temperature, anthropogenic pressure and increased behavioural alertness in summer.
Overall, integrating biologgers with machine learning provides a robust framework to identify welfare states in seabass, combining physiological, behavioural, and environmental data to inform management, optimise protocols, and enhance welfare-oriented aquaculture practices.

WELFARE Using behaviour and physiology to understand fish welfare

Behavioural and neurophysiological evidence for exercise choice as a welfare intervention in salmonids (Salmo salar and Oncorhynchus mykiss)

Bellio, Martina[1] (*martina.bellio0@nmbu.no*) Oral Presentation
Co-Author: Vindas, Marco A.[1]; Sneddon, Lynne [2]; Newberry, Ruth C. [1]
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Abstract: Behavioural choice paradigms provide a robust framework to identify positive welfare states when combined with physiological and neurochemical indicators of positive affective state. In mammals, voluntary exercise (i.e. exercise choice) has been associated with improved stress and pain resilience. Conversely, fish studies predominantly rely on forced exercise protocols, and comparable responses to exercise choice have not yet been elucidated. We investigated exercise choice in juvenile Atlantic salmon and rainbow trout using an arena that allowed movement between flow and calm zones and examined how this intervention alters stress- and pain-related behavioural and neurobiological responses. In a controlled experiment, we quantified spatial preference, plasma cortisol, and monoaminergic activity in forebrain regions involved in affective processing. Atlantic salmon preferred flow zones while intermittently returning to calm areas, indicating self-regulated exercise, and exhibited significantly lower plasma cortisol than controls. Neurochemical analyses revealed elevated dopamine and serotonin concentrations in the dorsomedial pallium of the exercise-choice group, the teleost functional equivalent of the mammalian amygdala. In a second experiment, we investigated how prior exercise (40-day

training) and post-stimulus exercise choice influence monoaminergic activity, plasma stress markers, and behavioural metrics in rainbow trout following an acute pain stimulus. Preliminary results indicate that fish allowed to exercise after pain displayed a more rapid behavioural recovery, whereas fish denied post-pain exercise exhibited behavioural and neurochemical profiles consistent with heightened arousal and potential frustration. Together, these findings indicate that exercise choice influences neurobiological processes associated with positive affect and improves recovery, highlighting potential welfare benefits of incorporating exercise choice into salmonid husbandry.

WELFARE Using behaviour and physiology to understand fish welfare

Enrichment and aggression in fishes: implications for animal welfare

Dijkstra, Peter *dijks1p@cmich.edu* Oral Presentation
Central Michigan University

Abstract: Aggression is a major welfare issue among many captive fish species. Physical enrichment can provide shelter and reduce the negative effects of aggression, but it may also increase territoriality if enrichments are perceived as valuable resources. In this talk, I will integrate insights from behavioral ecology and animal welfare science to present a conceptual framework for how physical enrichment shapes aggression and wellbeing in captive fish. Drawing on studies in highly territorial cichlid fish, I will highlight mechanisms through which enrichment mitigates aggression, including promoting conflict avoidance and redirecting motivation away from competition. Finally, I will discuss the need for more controlled experiments to isolate how specific enrichment attributes (e.g. size, complexity, placement) influence behavior, aggression, and welfare outcomes.

WELFARE Using behaviour and physiology to understand fish welfare

Why“stress” no longer serves us: retiring an ambiguous term to advance fish welfare

Currie, Suzie[1] (*suzie.currie@ubc.ca*) Oral Presentation
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Abstract: Fish health is routinely judged from behaviour interpreted alongside physiology, yet the umbrella term “stress” conflates stimuli, responses, and pathology, obscuring mechanism and hindering our interpretation of homeostasis. We advocate replacing “stress” in fish biology with the Reactive Scope Model (RSM; described by Romero and colleagues) as the primary interpretive framework for understanding responses to challenging environments. We study social interactions and temperature as distinct and combined challenges and use these to exemplify the RSM framework. The RSM defines functional ranges (i.e., predictive homeostasis, reactive homeostasis, and homeostatic overload/failure) for physiological mediators over time and explicitly links wear-and-tear accrued in the reactive range to reduced scope and increased risk of overload. Applying the RSM to understanding social interactions (e.g., social dominance hierarchies) in fishes

reveals consistent, mechanistic patterns. During agonistic encounters, both individuals transiently enter the reactive range; thereafter, subordinates often maintain elevated glucocorticoids and altered monoaminergic/neuropeptide signalling, accumulating wear-and-tear that suppresses growth and reproduction, thereby lowering reactive scope. In contrast, dominants typically return mediators (e.g., cortisol) to the predictive range and retain greater capacity to tolerate subsequent challenges. Importantly, this RSM framework explains why subordinates show reduced thermal tolerance, altered cardiorespiratory responses, and higher pollutant susceptibility, outcomes historically labelled as "social stress" effects but more precisely described as positions within this framework. Using environmental challenge (rather than "stressor") as any perturbation that shifts position along the RSM continuum clarifies mechanisms, standardizes language, and directly identifies levers, mediators, time, and accumulated wear-and-tear, yielding testable predictions for refinement in experimental biology.

WELFARE Using behaviour and physiology to understand fish welfare

Effect of fishing stress on reproductive output of wild cod

Jacson, Lea [1] (*Lea.jacson@outlook.fr*) Oral Presentation
Co-Author: Ásta Ólafsdóttir, Guðbjörg [1]; Sayyari, Amin [2]; Chavarie, Louise [2]; Vindas, Marco A.[3]
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Abstract: Welfare in wild-capture fisheries remains largely understudied and rarely incorporated into practice. A clearer understanding of what welfare means for wild fish, along with reliable ways to quantify and apply it, could offer substantial benefits in product quality and economic value, and is essential to ensuring ethical and sustainable practices. As Atlantic cod (Gadus morhua) stocks are declining faster than they can naturally recover, there is an urgent need for more sustainable harvesting practices. Time–area fishing closures during the cod spawning season have been implemented in Iceland and the Faroe Islands, restricting fishing activities. However, there is still a lack of understanding of how this measure affects the reproductive output of wild cod. Our objective is to characterize how fishing pressure (i.e. stress) may be affecting gamete quality and viability in wild cod. We hypothesize that reduced fishing activity will improve welfare and thereby enhance gamete quality, with potential relevance for long-term reproductive performance. To evaluate these differences, we will assess egg and sperm quality, measure cortisol levels in gametes, and monitor fertilization rate, embryonic deformities, developmental progression, and hatching success from samples collected during the spawning season in Iceland and The Faroe Islands. Furthermore, these parameters will be correlated to the parental neuroendocrine profile, to understand parental effects. Based on this approach, we expect that reduced fishing activity will improve welfare and thereby enhance gamete quality, thereby increasing sustainable fishing practices.

WELFARE Using behaviour and physiology to understand fish welfare

Turbine design shapes fish welfare: acute injury, mortality, and sublethal growth effects in juvenile white sturgeon

Zillig, Kenneth[1] (*kzillig@umn.edu*) Oral Presentation
Co-Authors: Baird, Sarah[2]; Watson, Sterling[3]; Lassar, Simone[3]; Schneider, Abraham[3] and Fangue, Nann[2]
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Abstract: Hydropower infrastructure poses significant risks to migratory and resident fish populations, with turbine blade strike representing a major source of direct injury and mortality. Conventional turbine runners characterized by thin, straight leading edges are known to cause severe injuries, yet few empirical comparisons exist between conventional and next-generation turbine designs. This study used juvenile white sturgeon (Acipenser transmontanus) to evaluate injury, mortality, and sublethal outcomes following passage through a model-scale turbine equipped with either a conventional runner or a fish-friendly runner. The fish-friendly runner, a Restoration Hydro Turbine (RHT), features thick, swept leading edges specifically designed to reduce the forces imparted to fish during blade strike. Both runners were tested across five blade peripheral speeds (15.0 to 27.6 m/s), with high-speed video documentation of all passage events. Fish assessed after passage through the conventional runner exhibited 42 to 78% survival at 48 hours, with approximately one-third of fish killed by blade strike. In contrast, the RHT achieved 100% immediate and 48-hour survival at all but the highest-speed condition (27.6 m/s; 95.6% survival). Following these acute comparisons, we evaluated the sublethal effects of RHT passage on post-passage growth. Fish that survived RHT passage exhibited modest but measurable reductions in growth relative to non-passage controls, indicating that even non-lethal passage events carry costs that persist beyond the immediate injury window. Together, these findings demonstrate that turbine design profoundly influences fish welfare outcomes, both lethal and sublethal, and support the case for incorporating fish-welfare criteria into the engineering and licensing of hydropower facilities.

WELFARE Using behaviour and physiology to understand fish welfare

Time-dependent effects of structural environmental enrichment on cognition and stress responses on juvenile European seabass

Rodríguez López, Noelia[1] (*nrodriguezlopez@imedea.uib-csic.es*) Oral Presentation
Co-Author: Arechavala-Lopez, Pablo[1]; Orgaz Ávalos, Alba María[1]; Hoyo Álvar, Esther[1]; Sáenz Lechón, Nicolás[2] and Gutiérrez Arriola, Juana María[2]
IMEDEA (CSIC-UIB), CITSEM-UPM

Abstract: Physical environmental enrichment (EE) is increasingly recognized as a strategy to enhance cognitive development and welfare in farmed fish by introducing structural complexity into rearing environments. This study evaluated the effects of structural EE on juvenile European seabass (Dicentrarchus labrax) reared in sea-cages under enriched (EE) and non-enriched (NE) conditions over 9 months. Cognitive, behavioural and physiological

stress responses were assessed after 3 months (medium-term) and 9 months (long-term) of exposure. Cognitive performance was evaluated using an emotional reactivity test and a maze test, complemented by physiological indicators. After 3 months, enriched fish showed enhanced cognitive performance and improved stress responses. In the emotional reactivity test, EE individuals travelled greater total distances and explored larger areas than NE fish, indicating increased exploratory behaviour and a greater capacity to cope with challenging situations. In the maze test, enriched fish solved the task earlier and improved performance more rapidly over time than NE fish. Together, behavioural and physiological results indicate that structural enrichment promoted functional cognitive enhancement and more adaptive stress responses at medium-term exposure. However, these positive effects were no longer detected after 9 months of continuous exposure. At this stage, cognitive performance and stress responsiveness did not differ between treatments, suggesting habituation to the enriched environment. The initial responses to structural novelty likely diminished over time, reducing its stimulatory value. Overall, the benefits of structural enrichment appear time-dependent and closely linked to environmental novelty. Incorporating variability into enrichment strategies may help prevent habituation and sustain long-term cognitive stimulation and welfare improvements in aquaculture systems.

WELFARE Using behaviour and physiology to understand fish welfare

Bringing together physiology, behavior, and cognition to understand the effect of environmental enrichment on positive welfare of farmed fish

Pettinau, Luca (*luca.pettinau@wur.nl*) Oral Presentation
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Abstract: Aquaculture is the world's fastest-growing food production sector, supplying ≈50% of fish consumed globally. As fish are sentient, show complex cognitive abilities, and likely feel pain in manners comparable to homeothermic vertebrates, welfare-oriented management in aquaculture is paramount. However, current aquaculture systems raise welfare concerns due to high stocking densities, lack of environmental complexity, handling stress, and limited opportunities for natural behaviours.Although these factors are widely assumed to be detrimental to welfare, experimental evidence of their impact remains limited. This is partly as welfare assessments rarely integrate behavioural and physiological measurements but rely on single performance indicators, which do not allow coherent welfare assessments. Farmed fish are often raised in barren systems designed for economic efficiency rather than welfare, causing chronic stress, impacting physiology and natural behaviour. Environmental Enrichment (EE) has been proposed asa potential strategy to improve positive welfare in aquaculture. By providing structural complexity or occupational stimulation,EE can promote natural behaviours, improve physiological performance, and reduce chronic stress. We hypothesise that EE could improve welfare by reducing chronic stress and shifting towards more robust phenotypes, expressed as improved aerobic scope, higher heart-rate-variability, increased thermal tolerance,

reduced anxiety-like behaviour, and ultimately a more positive state-of-mind. Our project will compare fish kept in enriched and barren environments and assess metabolic rate, cardiac performance, thermal tolerance, behavioural traits, and cognitive bias. By integrating physiological, behavioural, and cognitive indicators, we aim to increase our understanding of fish positive welfareand validate EE strategies to improve welfare and climate resilience in aquaculture.

WELFARE Using behaviour and physiology to understand fish welfare

The key importance of behavior and physiology to better understand fish welfare and resilience in aquaculture scenarios

Gesto, Manuel[1] (*mges@aqua.dtu.dk*) Oral Presentation
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Abstract: Ensuring high standards of fish welfare is increasingly recognized as a cornerstone for sustainable aquaculture, and the demand for robust welfare surveillance programs is particularly strong within the European Union. Current operational practices for welfare monitoring typically rely on Operational Welfare Indicators (OWIs), which are deliberately simple, easy to score, and practical for industry use. However, these indicators are often applied at the end of the production cycle or at specific critical stages, providing limited insight into the underlying processes that shape the animals' subjective experience and resilience.

A deeper understanding of welfare dynamics requires going beyond static, end-point assessments. Behavioral and physiological measures, such as activity patterns, feeding behavior, social interactions, stress endocrinology, and nutritional variables, offer essential information on the timing, severity, and recovery trajectories associated with different welfare risks. These metrics allow for a more precise interpretation of how fish perceive and cope with challenges, ultimately supporting better predictions of short- and long-term welfare outcomes.

In this presentation, our own experimental data will be integrated with findings from the literature to illustrate the relationships between commonly used OWIs and behavior- and physiology-based indicators in aquaculture-relevant scenarios. By examining these relationships, we highlight opportunities for refining welfare assessment frameworks to capture dynamic, process-oriented aspects of welfare and resilience. Our aim is to support the development of more informative and responsive welfare monitoring systems that align scientific understanding with practical implementation in modern aquaculture.

This work was co-funded by the European Union's Horizon Europe Project 101136346 EUPAHW

WELFARE Using behaviour and physiology to understand fish welfare

Disruption of the endocannabinoid system during gastrulation produces long-term behavioral and reproductive deficits in zebrafish.

Khan, M. D. Hasanuzzaman (*mdhasanu@ualberta.ca*) Oral Presentation
Co-Author: Ali, Declan
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Abstract: Cannabis is the most widely used recreational drug worldwide, and increasing use during pregnancy raises concerns about its potential developmental effects. The endocannabinoid system (eCS) plays a critical role in embryonic neurodevelopment, and its perturbation during early stages may result in long-lasting physiological and behavioral alterations. This study investigated the effects of brief embryonic exposure to Δ9-tetrahydrocannabinol (THC) and cannabidiol (CBD) on zebrafish (Danio rerio) development, swimming behavior, and reproductive physiology. Embryos were exposed to 0.01, 0.1, or 1.0 mg/L THC or CBD during gastrulation (0–24 hours post-fertilization) and assessed across larval and adult stages.

Larval zebrafish exposed to increasing concentrations of THC or CBD exhibited significantly decreased escape responses elicited by auditory/vibrational stimuli at 6 dpf compared to controls, indicating impaired sensorimotor function. In adulthood, embryonic THC exposure resulted in heightened locomotor and exploratory swimming behavior in open field (OPF) and novel object recognition (NOR) assays, as well as dose-dependent social deficits/reduced social motivation in shoaling behavior. Whereas, CBD exposure produced subtler long-term behavioral effects, though lower doses were associated with reduced social interaction. Additionally, preliminary findings indicate that embryonic THC exposure may impair reproductive health, as adult females exposed to 1.0 mg/L showed increased ovarian atretic oocytes (AO) and signs of accelerated reproductive decline compared to lower-dose groups and controls, warranting further histological investigation.

Collectively, these findings demonstrate that transient cannabinoid exposure during gastrulation induces persistent neurobehavioral and reproductive alterations in zebrafish, highlighting early embryonic sensitivity to endocannabinoid disruption and potential long-term risks of prenatal cannabinoid exposure.

WELFARE Using behaviour and physiology to understand fish welfare

Density and aggression in zebrafish: Resource Defense Theory explains conflicting results

Oldfield, Ronald[1] (*rgo.oldfield@gmail.com*) Oral Presentation
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Abstract: Zebrafish (Danio rerio) is a model organism important in biomedical research. Laboratories typically house zebrafish in small, barren containers, which results in high

densities of zebrafish per volume of water. Surprisingly, some studies have reported that zebrafish are more aggressive at higher densities, but other studies have reported that zebrafish are less aggressive at higher densities. To resolve the relationship between density and aggression, we observed zebrafish in volumes ranging from small, rack-mount laboratory containers to large glass aquaria. We quantified the behavior of male zebrafish, in male dyads (Experiment 1) and mixed sex trios (Experiment 2), in containers of different sizes that were either barren or enriched with gravel and live plants. Male zebrafish in 1-L, 3-L, and 10-L containers performed aggressive behaviors more frequently and spent less time swimming than they did in larger aquaria. The effects of physical enrichment were less apparent. We then graphed our results together with those obtained in previous studies. The density ranges tested in previous studies varied greatly: density values considered high in some studies were considered low in others. Consistent with Resource Defense Theory, our graph revealed an inverted U-shaped curve, in which zebrafish were most aggressive at intermediate densities, thereby resolving perceived inconsistencies. Although aggression is low at both high and low densities, reducing aggression by stocking fish at low densities in large containers is better than high densities in small containers because space limitation prevents locomotory behavior, which has known benefits in fishes.

WELFARE Using behaviour and physiology to understand fish welfare

Improving the stress response and welfare of Nile tilapia, *Oreochromis niloticus* (Linnaeus, 1758) through environmental enrichment

Campang, Joseph (*j.d.g.campang.2361300@swansea.ac.uk*) Oral Presentation
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Abstract: Nile tilapia (Oreochromis niloticus) is one of the most widely farmed fish globally, known for its adaptability, rapid growth, and significant contribution to food security. The stress response of Nile tilapia has received much attention due to its importance for health, productivity, and sustainability, but relatively little is known about how to improve its welfare. We tested the effects of integral enrichment on the stress response and welfare of Nile tilapia through the juvenile phase. Integral enrichment was achieved through the provision of refuges and artificial plants (structural enrichment), in-tank currents (occupational enrichment), and dietary supplementation (nutritional enrichment). After the rearing period, a multi-variable welfare response was computed based on survival, growth, condition factor, opercular beat rate (OBR), boldness, exploratory behaviour, plasma and faecal cortisol, and operational welfare indicators (OWIs). The results showed that fish reared under enriched conditions recover more rapidly from handling stress than those reared under barren conditions, suggesting a greater ability to restore homeostasis. Our study indicates that integral enrichment can improve the welfare of Nile tilapia, although further research is required to identify the most useful suite of interventions.

WELFARE Using behaviour and physiology to understand fish welfare

From behavior to biomass: early enrichment enhances trout welfare and performance

Jonassen, Ane (*ane.kjelle.jonassen@nmbu.no*) Oral Presentation
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Abstract: Intensive aquaculture production challenges fish health and welfare, affecting both economic and ethical sustainability. Increasing environmental complexity (environmental enrichment, EE) has been shown to improve fish physiology and welfare and is assumed to promote natural, adaptive behaviors. This study investigated how early exposure to environmental complexity affects growth and welfare-related behavioral responses in juvenile rainbow trout. After 90 days of rearing in EE or barren environments (BE), fish were transferred to individual isolation for two weeks to assess stress coping ability. No differences in feed intake was observed between the treatments, but mass gain was more than twice as high in fish transferred from enriched compared to barren rearing. EE fish also showed significantly lower frequencies of stereotypic vertical swimming after transfer to the novel environment, suggesting greater behavioral flexibility and resilience. The increased occurrence of stereotypic behavior in BE fish, emerging only after exposure to the novel environment, supports the conclusion that growth differences are influenced by behavioral correlates of stress coping ability in EE- and BE-reared fish. These results point to the value of broadening behavioral indicators used in aquaculture, as behavioral outcomes of welfare translate, to some degree, directly to growth performance following environmental change. This observation should serve as a strong motivation to maintain a clear focus on using behavior to assess welfare status in aquaculture production.

WELFARE Using behaviour and physiology to understand fish welfare

Enrichment, emotions and fish welfare - integrating behavioural measures to provide better fish care

Rose, Paul (*p.rose@exeter.ac.uk*) Oral Presentation
University of Exeter, Exeter

Abstract: Animal welfare consists of physical, behavioural, and psychological components. These can all be assessed to improve our understanding of how animals under human care respond to the resources and environments provided. Fish welfare in zoological collections and in home aquaria is becoming an increasing topic of interest. Quantifying physical and behavioural responses to housing and husbandry (including welfare-positive elements of husbandry, such as environmental enrichment) is possible in small scale (e.g., domestic aquariums) and in larger, complex zoological exhibits. Application of enrichment workflows (where enrichment is created based on a behavioural output that a caregiver would like their fish to perform) provide further opportunities for control and choice, which enable positive welfare outcomes to be experienced. This talk will explain the concept of enrichment workflows and link evaluation of their efficacy to Qualitative Behavioural Assessment (QBA) - a method that uses behavioural expression ("body language") to infer

the psychological component of animal welfare. QBA will be explored to show how a fish's individual responses to human-provided care can be objectively monitored via a specific list of descriptors of behavioural expression, supported by assessment of physical characteristics. Finally, an example that integrates behavioural ecology, social system and environmental heterogeneity will be used to emphasise the importance of species-specific enrichment plans and how enrichment used for fish (as with other taxa) should be based on appropriate evidence to ensure that husbandry changes are valid, welfare-positive and biologically relevant.

WELFARE Using behaviour and physiology to understand fish welfare

Stress coping, allostasis, and the welfare of fish in aquaculture

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Norwegian University of Life Sciences

Abstract: Ensuring good fish welfare in aquaculture is essential not only for ethically sound and sustainable production, but also for effective disease prevention. Close monitoring and recording of operational welfare indicators (WI's) is therefore a critical component of aquaculture practices, typically relying on scoring based on established protocols. A consensus definition of animal welfare is "the individual's subjective experience of its mental and physical state as a consequence of its attempts to cope with its environment." Under this definition, the relevance of any specific indicator cannot be directly validated, as subjective experience is not accessible to scientific measurement. Nevertheless, complex phenomena such as emotional and cognitive function are grounded in measurable neurobiological processes, i.e. communication between neurons in a network. Furthermore, the principle of allostasis recognizes that functional stress responses are essential for coping with changing environments, and that negative welfare consequences emerge only when prolonged or severe challenges exceed the animal's capacity to cope. Recent results will be reviewed, showing that compromised welfare is associated with a distinct neurobiological signature characterized by reduced responsiveness of central regulatory systems, including brain serotonergic signalling and the HPI axis. Notably, across several environments, fish in poorer welfare condition exhibit a markedly blunted serotonergic response to acute stress compared with individuals in better condition. Taken together, these findings outline an approach for verifying the biological relevance of WI's suitable for operational monitoring, enabling welfare data to be used more effectively for documentation, decision support, and research.

WELFARE Using behaviour and physiology to understand fish welfare

Improving fish welfare in the ornamental trade

Sloman, Katherine (*katherine.sloman@uws.ac.uk*) Oral Presentation
University of West Scotland

Abstract: Fish within the ornamental trade experience cumulative stressors over multiple transport stages which can affect their welfare. In this talk I will present work from my research group looking at different stages in the transport chain to identify key welfare concerns and potential interventions. Working with the ornamental trade, we have documented the water quality fish experience through international supply chains, looking at both the shipping of ornamentals from aquaculture in Singapore, and transport of wild caught ornamentals in the Amazon. Our results show that even when water quality parameters remain within acceptable ranges, fishes still experience abrupt changes in water chemistry which represent important welfare considerations. To improve welfare, we have investigated potential enhancements during transport including benefits of adding physical enrichment to transport bags. We have also found potential welfare benefits of adding waterborne cannabidiol during transport. Following transport, on arrival at a retailer, the social environment a fish is added to will also influence their welfare. Fish welfare within the final destination of a home aquarium is probably the least well researched scientifically; here we have explored the reasons why home aquarists keep substrate in their tanks and the influence of substrate on tank microbiome. Finally, we are considering how we should monitor fish welfare in the ornamental trade and the potential to utilize welfare tools designed for food-fish aquaculture.

WELFARE Using behaviour and physiology to understand fish welfare

Stunning challenges: Operational indicators flag failures, but neurological validation is needed to confirm stunning effectiveness in seabass and seabream

Saraiva, Joao[1] (*joao@fishethogroup.net*) Oral Presentation
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[1] Fish Etho Group Association; [2] Swedish University of Agricultural Sciences, [3]IMEDEA (CSIC-UIB), [4]University of Padova, [5]University of Innsbruck

Abstract: Humane slaughter in aquaculture depends on accurate assessments of insensibility, yet commonly used operational indicators remain poorly validated against neurological benchmarks. This study compared operational indicators such as the loss or recovery of equilibrium, 'eye-roll' reflex, and ventilation reflex with visually evoked responses (VERs) in European seabass (Dicentrarchus labrax) and gilthead seabream (Sparus aurata) subjected to anaesthetic immersion or in-water electrical stunning. Operational indicators consistently preceded or lagged behind VERs, risking misjudgment of fish sensibility. Notable species differences emerged, with seabass losing and recovering indicators faster than seabream, whose indicator sequences varied depending on stunning method. Among all indicators, the ventilation reflex aligned most closely with

VERs in both species, suggesting it may be the most reliable operational proxy. Despite the increasing use of electrical stunning in commercial settings, rapid recovery times (within seconds to minutes) observed in most fish raise serious welfare concerns. These findings underscore the urgent need to improve and validate electrical stunning methods for seabass and seabream before they can be considered humane and are widely implemented. Until neurological tools are feasible for commercial use, thorough species- and method-specific validation of operational indicators remains essential for safeguarding fish welfare.

WELFARE Using behaviour and physiology to understand fish welfare

Post-release welfare in marine recreational fisheries: integrating angling variables with physiological biomarkers in the two-banded seabream Diplodus vulgaris

Guerreiro, Pedro M[1] (*pmgg@ualg.pt*) Poster Presentation
Co-Author: Nuñez-Velazquez, Samira[1]; Jacinto, Gonçalo[2]; Rangel, Mafalda[1]; Veiga, Pedro[1]; Pontes, João[1]; Saraiva, Joao[3] and Costa, Rita A.[1]
[1]Centre of Marine Sciences CCMAR/CIMAR LA, [2]University of Algarve, [3]Fish Etho Group Association

Abstract: Catch-and-release is widely promoted in recreational fisheries, but its conservation value depends on how rapidly fish recover from capture stress. We examined post-release physiological responses in two-banded seabream, Diplodus vulgaris, by integrating two complementary datasets to test how recovery time and capture-related variables shape stress biomarkers after boat angling. Fish were collected, processed, and held in a floating sea cage for recovery periods of 4–6 h or 26–28 h. Individuals were sampled either immediately after capture and again after holding, or only after holding, and blood glucose, lactate, osmolarity, and cortisol were quantified. We also evaluated the effects of hook location, hook size, fight time, air exposure, and body mass.
Recovery was not uniform across biomarkers. Longer confinement was associated with lower lactate, consistent with partial metabolic recovery, but glucose increased with cage time rather than declining over the observed recovery period. Osmolarity showed weaker overall recovery patterns, while cortisol responses were more complex and, after baseline adjustment, were associated with cage time, hook location, hook size, and body mass. Among all predictors, hook location emerged as the clearest determinant of post-release condition: swallowed hooks were associated with mortality or poorer physiological status after holding, particularly higher lactate and osmolarity. By contrast, fight time and air exposure had comparatively weak or inconsistent effects under the conditions tested. Together, these results show that post-release recovery in D. vulgaris is biomarker-specific rather than uniform across physiological systems and is strongly shaped by capture characteristics, especially hook location. Reducing deep hooking may therefore be one of the most effective ways to improve fish welfare and strengthen the biological credibility of catch-and-release in recreational angling.

WELFARE Using behaviour and physiology to understand fish welfare

Links between forebrain monoaminergic activity and operational welfare indicators in farmed Atlantic salmon (Salmo salar)

Gundersen, Hannah N.[1] (*hannah.normann.gundersen@nmbu.no*) Poster Presentation
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Abstract: Farmed Atlantic salmon represent a major global aquaculture industry; however, high mortality rates highlight persistent welfare challenges. Despite advances in health monitoring, welfare assessments still rely heavily on external operational welfare indicators (OWIs), and it remains uncertain to what degree these outward signs reflect actual stress coping ability and underlying neurobiological state. In this context, we examined whether commonly used OWIs correspond to the responsiveness of key allostatic regulatory mechanisms, including the hypothalamus–pituitary–interrenal (HPI) axis and central monoaminergic signalling, in adult Atlantic salmon (Salmo salar) reared in seawater. As predicted, stress exposure (handling and overnight confinement in low water volumes) led to elevated plasma cortisol and increased monoaminergic turnover in both the brainstem and telencephalon.

Most external indicator scores increased during the stress test, with the post-stress score averaging 2.21 times the pre-stress score. The greatest contributors to the OWI increase were eye damage (3.5 fold change) and skin bleeding (2.6 fold change).

The combined OWI score also increased significantly. Significant positive correlations emerged between OWI score and telencephalon dopamine and DOPAC concentrations, as well as a negative correlation with telencephalon 5-HIAA. However, all relationships showed significant interaction with stress treatment; strong associations were present in control fish, whilst these associations were attenuated in stressed fish. No relationship was found between OWI score and plasma cortisol. These results suggest that external damage, measured by OWI scores, is associated with monoaminergic tone and turnover in the telencephalon under baseline conditions, but that acute stress disrupts this relationship. In this context, we examined whether commonly used OWIs correspond to the responsiveness of key allostatic regulatory mechanisms, including the hypothalamus–pituitary–interrenal(HPI) axis and the central serotonergicsignallingsystem, in adult Atlantic salmon (Salmosalar) reared in seawater. As predicted, stress exposure (handling and overnight confinement in low water volumes) led to elevated plasma cortisol and increased serotonergic turnover in both the brainstem and telencephalon.

Individually, external welfare indicators such as fin damage, scale loss, and wounds showed little or no relationship to physiological stress-coping capacity. In contrast, when combined into a total OWI score, a significant negative correlationemergedwith brainstem 5-HT turnover. This suggests that reduced welfare status is reflected in a diminished capacity of this centralsignallingsystem to adapt to new stressors. In other words, cumulative welfare impairment—rather than any single external indicator—captures

biologically relevant variation in brain functions involved in behavioural control as well as cognitive and emotionalprocessing.

WELFARE Using behaviour and physiology to understand fish welfare

Effects of feeding time on diurnal expression of clock genes and lipid metabolism-related genes in the brain and peripheral tissues of Malabar grouper (Epinephelus malabaricus)

Lu, Siqi[1] (*lusiqi85937960@gmail.com*) Poster Presentation
Co-Authors: Fukunaga, Kodai[1]; Zhu, Yafan[2]; Takekata, Hiroki[1] and Takemura, Akihiro[1]
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Abstract: Feeding time is a strong synchronizer of peripheral clocks and metabolic processes in vertebrates. However, its effects of lipid metabolism in marine fish remain unclear. This study investigated the effects of feeding time on the diurnal expression of clock genes and lipid metabolism–related genes in the central nervous system tissue (diencephalon) and peripheral tissues (liver and intestine) of juvenile Malabar grouper (Epinephelus malabaricus). Fish were assigned to unfed, 08:00-fed, and 14:00-fed groups. Gene expressions in the diencephalon, liver, and intestine were analyzed over a 24 h cycle. In the unfed group, clock genes exhibited clear diurnal rhythms in all tissues, with higher expression during the light phase. Per2 peaked in the early light phase, while other clock genes peaked in the late. In the liver, several lipid metabolisms–related genes, including Srebf1, Ppary, Fas, Dgat2, Cpt1, Hsl1, and Lpl1, showed significant rhythmicity. Feeding time did not alter the phase of clock gene expression in the diencephalon, indicating stability of the central clock. In contrast, feeding induced phase delays and disrupted rhythmicity in peripheral tissues. In the liver, clock gene expression was delayed by 1–3 h, with greater disruption in the 14:00-fed group. The intestine was more sensitive, showing larger phase delays and loss of rhythmicity in Bmal1 and Bmal2. Feeding also abolished rhythmicity of Srebf1 and Lpl1 and delayed Cpt1 expression. These results demonstrate that feeding time acts as a potent zeitgeber for peripheral clocks and lipid metabolism in this species, suggesting feeding schedules should consider circadian physiology to optimize metabolic regulation in aquaculture.

WELFARE Using behaviour and physiology to understand fish welfare

Stress reactions in rainbow trout (Oncorhynchus mykiss) reared in enriched vs. barren environments underline the importance of behavioural observations for assessing coping ability and welfare status of fish

Olsen Methlie Wedaa, Louise (*louise.wedaa@gmail.com*) Poster Presentation
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Norwegian University of Life Sciences

Abstract: Environmental enrichment is widely explored as a strategy to improve welfare in captive fish. Recent results from our group on farmed rainbow trout (Oncorhynchus mykiss)indicate that early environmental conditions influence growth, brain morphology and behavioural responses to environmental novelty. Here, we examined whether early-life exposure to either barren (BE) or structurally enriched environment (EE) induces lasting changes in stress responsiveness. Following transfer to a novel environment and a 12-day period of social isolation,EE fish exhibited significantly higher specific growth rate than control (BE) fish. To assess whether these growth differences reflect broader variation in behavioural syndromes or coping styles, the same individuals were subsequently subjected to a standardized 24-hour confinement stress test. While indicators of HPI-axis activation (circulating plasma cortisol) and post-stress brain monoaminergic activity (serotonin, dopamine and norepinephrine) did not differ between treatments, marked behavioral differences emerged.BE fish spent a substantially higher proportion of time moving than EE fish, both initially (31.85 % vs 16.65 %) and after 23 hours (20.4 % vs 9.07 %). Locomotor activity under acute stress is widely regarded as a behavioural correlate of stress coping style in fish.
These results demonstrate persistent behavioural divergence despite the absence of detectable physiological differences after 24 h of stress. Current welfare assessments in aquaculture rely heavily on scoring external operational indicators, together with physiology and health measures. In this context, the above result highlights the importance ofalso incorporating behavioural observations when evaluating coping ability and welfare status in farmed fish.

WELFARE Using behaviour and physiology to understand fish welfare

Behavioural responses of gilthead seabream (Sparus aurata) to the administration of painful stimuli

Saraivo, Joao[1] (*joao@fishethogroup.net*) Poster Presentation

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Pain is an aversive sensory and emotional experience whose investigation in fishes remains limited. In the present study, we evaluated the effects of acetic acid (AA) into the lips of gilthead seabream (Sparus aurata), an important aquaculture species. Two experiments were carried out in which fish were subcutaneously injected in the lips. In Experiment 1, fish received sham handling (needle insertion), PBS (control solution), 1, 5 or 10% AA. In Experiment 2, fish received PBS, 10% AA, or 10% AA combined with lidocaine at 1 or 2 mg mL-1. In both experiments, behaviour was recorded 1 h before treatment (Time 0) and at 0.5, 1.5, 3 and 6 h post-injection (p.i.). Qualitative inspection of the lips showed that 10% AA was associated with visible swelling and mouth opening movements, which were not

observed in controls. 10% AA increased freezing, with greater use of the lower zone of the tank at 0.5, 1.5 and 3 h p.i. Co-administration of lidocaine reversed these behavioural changes at both doses tested. These results are in line with the responses found in other species to painful stimuli and suggest that 1) AA elicits a typical pain-like response in gilthead seabream and 2) the physiological sensing component shares the same conserved mechanisms of reception and transmission of painful signals as other vertebrates. These results add to the mounting evidence of pain in fishes and have ethical implications on the treatment of this (and other) fish species under a welfare perspective.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish: A tribute symposium to Dr. Frederick (Rick) W. Goetz

From reproductive physiology to molecular physiological ecology of fish: A tribute symposium to Dr. Frederick (Rick) W. Goetz

Josep Planas, Julien Bobe and Simon MacKenzie

Abstract: This symposium is to pay tribute to Dr. Frederick (Rick) W. Goetz and his long and outstanding career in fish biology. Rick has made seminal contributions to a number of fields in fish biology, including reproductive physiology, immunology, growth and physiological ecology. Importantly, Rick has been a pioneer in the development and application of molecular biology and genomic approaches to the study of fish biology. Throughout his full-time tenures at the University of Notre Dame in Notre Dame, IN, Woods Hole Oceanographic Institution in Woods Hole, MA, the University of Wisconsin in Milwaukee, WI, and lastly at the National Oceanographic Atmospheric Administration in Seattle, WA, Rick mentored and collaborated with many young and established scientists who have benefited from his inexhaustible energy, love for fish biology, dedication to rigorous research, and friendship. The proposed symposium will bring together many of his past and more recent students and collaborators to celebrate his career and commitment to the advancement of fish biology.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

Introduction to the symposium "From reproductive physiology to molecular physiological ecology of fish": a tribute to Dr. Rick Goetz

Planas, Josep
International Pacific Halibut Commission

Abstract: An introduction to the topic of the Symposium "From reproductive physiology to molecular physiological ecology of fish: a tribute to Dr. Rick Goetz" will be given by the organizers.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

Assessing heritable phenotypic variation in lake charr ecomorphs of Lake Superior

Goetz, Rick - Oral Presentation
University of Wisconsin

Abstract: Resource polymorphisms are common among charr (Salvelinus) ecomorphs. In North America, there are lake charr (Salvelinus namaycush) ecomorphs exhibiting variations in morphology, physiology and behavior. Many of these occur in deep lake

ecosystems (e.g., Lake Superior, Great Bear and Slave Lakes) where depth provides a variety of habitats varying in temperature, pressure, and forage composition. Which phenotypic variations are heritable or result from environmental plasticity is a fundamental question for understanding the sympatric evolution of these ecomorphs and may have profound impacts on their resiliency to habitat change in the future. In Lake Superior, there have been four reported ecomorphs; leans, siscowets, redfins and humpers. However, based on observations particularly in complex bathymetric habitats (e.g., Isle Royale, Stannard Rock, Klondike Reef), there are certainly more ecomorphs that have yet to be described. So far, ecomorphs (particularly leans and siscowets) can be differentiated based on morphometry, growth, habitat, incidence of skip spawning, pyloric caeca, and to a lesser extent diet. They also exhibit physiological differences in the amount of lipid and glycogen stored in the muscle and liver as well as in the effects of lamprey wounding. This talk will synthesize efforts that have been made to delineate which of these phenotypes have a genetic basis and to what extent. It relies heavily on the results of a 15-year, multi-generational common garden rearing experiment of lean and siscowet lake charr.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

Linking Molecular Responses to Population Consequences: Differential Effects of Sea Lamprey Parasitism in Siscowet and Lean Lake Trout

Murphy, Cheryl[1] (*camurphy@msu.edu*) Oral Presentation
Co-Author: Firkus, Tyler[2]; Alderman, Sarah[3]; Goetz, Rick[2]; Ivan, Lori[1]; Sitar, Shawn[4]; Smith, Sara[5] and Lika, Konstadia[6]
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Abstract: Rick Goetz's work on lake trout reproductive physiology, lipid metabolism and ecotype differentiation laid the groundwork for understanding how physiology shapes life-history strategy. Building on his common-environment rearing of siscowet and lean lake trout, we investigated how parasitism alters molecular and energetic pathways in these contrasting ecotypes and how those changes scale to individual and population outcomes. Using transcriptomic and endocrine analyses, we identified ecotype-specific responses to sea lamprey parasitism. Parasitism was associated with shifts in steroidogenic pathways, steroid binding proteins, and lipid metabolism genes, accompanied by changes in circulating steroid concentrations and energy storage. These responses differed between siscowet and lean trout, consistent with their distinct lipid allocation strategies and growth trajectories. To determine how these molecular and physiological shifts translate into performance, we incorporated empirical measurements into Dynamic Energy Budget (DEB) models. Simulations captured the parasitism effects - that energy allocation to maintenance, growth, and reproduction is different in the two ecotypes when faced with parasitism stress with implications for lifetime reproductive output. We then embedded individual-level energy dynamics into an individual-based population model to explore long-term consequences under varying parasitism pressures and with one or both ecotypes present. Model projections indicate that ecotype-specific energetic strategies in

response to parastism. mediate population dynamics. In tribute to Rick Goetz, this work reflects a systems-oriented approach he championed—connecting molecular physiology, energetics, and life-history strategy to better understand the dynamics of Great Lakes lake trout populations and how they respond to stressors like sea lamprey parasitism.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

Gene Presence-Absence Variation and DNA Methylation Landscapes Underlying Ecotype Divergence in Lake Trout (Salvelinus namaycush)

Roberts, Steven [1] (*sr320@uw.edu*) Oral Presentation
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Abstract: Lake trout (Salvelinus namaycush) exhibit pronounced ecotypic diversification across North American lakes, including sympatric morphs (lean, siscowet, humper, redfin) that differ in depth distribution, lipid metabolism, buoyancy regulation, and life-history strategy. While most genomic studies in this system have emphasized SNP variation, the roles of structural genomic variation and epigenetic regulation in ecotype divergence remain largely unexplored. Here, we integrate pangenome analysis and whole-genome DNA methylation profiling to examine how gene presence--absence variation (PAV) and epigenomic state jointly contribute to ecological specialization. Using high-coverage long-read genome assemblies and population-scale resequencing, we construct a lake trout pangenome to identify gene gains and losses across ecotypes. In parallel, we characterize DNA methylation landscapes and identify differentially methylated regions (DMRs) associated with ecotypes. We detect substantial gene content variation among ecotypes, with PAVs enriched in gene families related to lipid transport, mitochondrial function, immune response, and sensory biology. Notably, ecotype-specific genes frequently exhibit distinct methylation signatures, and DMRs are enriched near PAV boundaries and in regulatory regions of metabolic genes. Our findings support a model in which adaptive divergence in postglacial fishes involves not only sequence-level variation but dynamic shifts in gene repertoire and epigenomic regulation. By integrating structural genomics with DNA methylation state, this work highlights how gene presence and gene regulation interact to generate ecological specialization in salmonids and advances a more comprehensive framework for understanding genomic mechanisms of adaptive radiation.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

Phenotypic and genotypic variability of Lake Char sharing the same spawning reef complex

Euclide, Peter[1] (*peuclide@iu.edu*) Oral Presentation
Co-Author: Moran, Max[1]; Hook, Tomas[2]; Seider, Mike[3]; Sitar, Shawn[4] and Goetz, Rick[5]
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Abstract: Lake Char (Salvelinus namycush) have evolved distinct ecomorphs (ecotypes) in sympatry multiple times without clear evidence of reproductive isolation. In Lake Superior, there are four described ecomorphs of Lake Char, each with distinct, but overlapping phenotypic traits and ecological niches. During most of the year, ecomorphs forage in different habitats and exhibit different feeding strategies. However, neutral genetic structure among Lake Char ecomorphs is lower than among spawning reefs, suggesting that there may be significant gene flow among ecomorphs. Furthermore, multiple ecomorphs can be found aggregating to spawn on or near the same spawning reef complexes suggesting physical reproductive isolation is limited. On Klondike Reef in eastern Lake Superior, all four ecomorphs were identified during the 2023 and 2024 spawning season. We confirm that there are significant phenotypic and dietary variation among identified ecomorphs using histology, morphometrics, stable isotopes, and fatty acids. Then we evaluate whole-genome level variation to attempt to determine the genetic mechanisms that underlie the phenotypic variability of lake char spawning on Klondike Reef in sympatry.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

Genomic evidence supporting panmixia of Pacific halibut in the Northeast Pacific Ocean

Jasonowicz, Andy (*andy.jasonowicz@iphc.int*) Oral Presentation
Co-Authors: Planas, Josep V.
International Pacific Halibut Commission

Abstract: The Pacific halibut (Hippoglossus stenolepis) is a large member of the Pleuronectidae family that is widely distributed in the North Pacific Ocean. The primary objective of the present study is to advance our understanding of Pacific halibut population structure in the Northeast Pacific Ocean. We have generated genomic sequences for 731 Pacific halibut collected during the winter spawning season from five wide geographic areas in the Northeast Pacific Ocean using low-coverage whole genome resequencing, a method that allows the characterization of genomic variation at very high resolution. Following alignment to the Pacific halibut reference genome, we observed average sequencing depths of ~3x and identified millions of single nucleotide polymorphisms (SNPs) throughout the Pacific halibut genome. Principal component analysis and unsupervised clustering were used to characterize population structure using approximately 3.7 million autosomal SNPs. Additionally, we conducted assignment testing

using a subset of these SNPs to assign individuals back to the spawning groups they were sampled from and used cross-validation to evaluate assignment accuracy. Our results identify a single genetic cluster of Pacific halibut in the Northeast Pacific Ocean despite the large spatial scale of sampling. Furthermore, we observed low assignment accuracies (<40%) when tracing individuals back to their population of origin. These results suggest the presence of a single panmictic genetic group of Pacific halibut in the Northeast Pacific Ocean that could be attributed to considerable movement and geneflow within this ocean basin.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

The influence of Sea Lamprey parasitism on Lake Charr reproductive physiology

Firkus, Tyler (*tyfirkus@uwsp.edu*) Oral Presentation
University of Wisconsin-Stevens Point

Abstract: Lake Charr populations in the Great Lakes faced significant declines due in part to the introduction of non-native Sea Lamprey. An estimated 45-75% of lake trout survive a sea lamprey parasitism event, but we have relatively little information about the physiological response of surviving Lake Charr. In a series of laboratory-based parasitism trials, we examined the sub-lethal effects of sea lamprey parasitism on a number of reproductive, immune, and growth endpoints. These studies identified critical consequences of Sea Lamprey Parasitism for reproductive performance, wound healing, protein and gene expression, and energy storage. Parasitized male lake trout from both morphotypes displayed reduced sperm concentrations one year following parasitism.Siscowet females with A-type wounds had reduced egg production compared with controls or females with B-type wounds, while parasitism status did not affect egg production for lean females.Though these results are preliminary, they suggest sea lamprey parasitism induced reproductive effects can have long term negative consequences for lake trout populations.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

Prostaglandin F2a is metabolized in taxon-specific manners demonstrating that this lipid may commonly serve as a precursor for species-specific spawning pheromones in ovulated fishes

Sorensen, Peter (*soren003@umn.edu*) Oral Presentation
Co-Authors: Brash, Alan
Vanderbilt University

Abstract: Nearly 40 years ago, Sorensen and colleagues working with Rick Goetz discovered that prostaglandin F2a (PGF2a), a lipid hormone that drives female sexual behavior in goldfish, is metabolized into at least 5 products and then released to function as a two-component spawning pheromone. Since that discovery, dozens of other species fish have been shown to detect waterborne prostaglandin F2a, suggesting that they too use this product and its metabolites as pheromones.However, the complete identity of this

pheromone and how PGF2a metabolites (PGFMs) might have species-specific function is not clear.In this study we injected goldfish with radiolabeled PGF2a, extracted the products, and used radio-chromatography to characterize and isolate these products which were then identified using mass chromatography.In addition, to PGF2a, we found that goldfish released 15keto-prostaglandinF2a as well as 13,14-dihydro-15keto-prostaglandin F2a and several novel glucose conjugates of these products.Additionally, we found that only unconjugated PGFs are detected by the male goldfish olfactory system showing that olfactory detection is highly specific, suggesting that differences in PGF2a metabolism could make pheromones species-specific.This possibility was supported by a follow-up study of PGF2a metabolism in 5 other species of fish which found that they released different suites of PGFMs showing that novel mixtures of PGFMs could function as species-specific pheromones.The identities and actions of these PGFMs await identification.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

Circulating microRNAs reveal egg-brain crosstalk and a brain-specific microRNA linked to the onset of the next reproductive cycle in iteroparous salmonids

Bobe, Julien (*julien.bobe@inrae.fr*) Oral Presentation
Institut National de Recherche pour l'Agriculture, l'Alimentation et l'Environment

Abstract: Mechanisms regulating the transition between consecutive reproductive cycles remain poorly understood, largely because they involve complex interactions between the ovary and the central nervous system. In rainbow trout (Oncorhynchus mykiss), an iteroparous species that spawns annually, removal of retained eggs from the body cavity was used as a physiological switch to trigger the onset of the next reproductive cycle. Circulating miRNA (c-miRNA) levels in blood plasma and ovarian fluid were monitored over time. Egg removal induced a marked downregulation of a single plasma c-miRNA, miR-139-5p, which is predominantly expressed in the brain. In contrast, distinct plasma c-miRNA profiles were observed when eggs were retained. Among the plasma c-miRNAs dynamically regulated under egg retention, miR-135c is highly expressed in the brain and pituitary, whereas miR-457a is predominantly expressed in the postovulatory ovary. Egg retention also triggered a pronounced decrease in ovarian fluid levels of miR-202-5p, a miRNA known to regulate egg production in fish, highlighting its potential as a non invasive biomarker of egg quality. These findings indicate that the transition between successive reproductive cycles involves bidirectional communication between retained eggs and the central nervous system. Notably, miR-139-5p emerges as a key candidate associated with initiation of the next reproductive cycle. We identified potential functional targets of miR-139-5p in rainbow trout and other iteroparous species that appear absent in semelparous salmonids, highlighting post-transcriptional regulation in the brain as a promising mechanism controlling reproductive cycling.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

Mediating Role of Thyroid Hormone in the Regulation of Temperature-Induced Changes in Early Gonadal Differentiation in Zebrafish.

Tailor, Nurulain (*nurulain.tailor1@ucalgary.ca*) Oral Presentation
Co-Authors: Habibi, Dr. Hamid
University of Calgary

Abstract: Sex determination and gonadal differentiation in fish are highly plastic processes influenced by genetic, environmental, endocrine, and molecular factors. Environmental variables, including temperature, can alter endocrine signaling and thereby affect sex differentiation. Thyroid hormones (THs) have been implicated in this process. Hypothyroid conditions bias sex ratios toward females, and hyperthyroid conditions favor males. However, whether thyroid hormones mediate temperature-induced effects on early gonadal differentiation remains unclear. This study tested the hypothesis that "thyroid hormone signaling mediates temperature-driven changes in early gonadal differentiation in zebrafish". Three experimental groups were established: euthyroid controls, hyperthyroid fish treated with thyroxine (T4), and hypothyroid fish exposed to methimazole and perchlorate that inhibit thyroid hormone synthesis. Treatments began at 3 days post-fertilization (dpf) and continued until adulthood (120 dpf). Fish were maintained at 28°C throughout development or exposed to an elevated temperature (35°C). Results demonstrated a significant increase in male development at 35°C compared to 28°C. Hypothyroid fish exhibited an altered pattern of gonadal differentiation and temperature-mediated response. Accompanying changes were observed in gonadal and thyroid tissues, growth, and gene expression patterns. The findings provide novel insights into how thyroid hormones and temperature influence gonadal differentiation, and the role of endocrine and environmental factors influencing sex differentiation in zebrafish.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

The integrated role of arginine vasotocin in zebrafish spermatogenesis

Habibi, Hamid R (*habibi@ucalgary.ca*) Oral Presentation
Co-Authors: Zanardini, Maya
University of Calgary

Abstract: Arginine vasotocin (AVT) is a neurohypophysial nonapeptide traditionally linked to osmoregulation and social behavior in fish. Although previous research has shown the presence of AVT receptors in both male and female gonads, information on the role of this nonapeptide in mediating gonadal function is limited. This study explores the function of AVT in the paracrine control of zebrafish spermatogenesis. By integrating ex vivo and in vivo approaches, we investigated AVT's direct effects on steroidogenesis and germ cell proliferation, as well as its interplay with gonadotropins. Transcript and protein analyses verified expression of avt and receptors (avpr1aa, avpr1ab, avpr2aa, avpr2ab, avpr2l) in zebrafish testis, supporting autocrine/paracrine roles. In addition, AVT stimulated basal androgen-driven spermiogenesis but suppressed early type B spermatogonial proliferation

in testes incubated for 7 days. In the presence of gonadotropins, AVT amplified FSH-supported expansion of undifferentiated (Aund) and type B spermatogonia while dampening FSH-induced 11-ketotestosterone (11-KT) output and Leydig cell factors. Conversely, AVT boosted LH-driven pre-meiotic and meiotic proliferation without affecting the final stages of spermiogenesis. Prolonged AVT treatment, in vivo, led to a dose-dependent accumulation of type A undifferentiated spermatogonia, reduced spermatozoa, and lowered plasma 11-KT. These changes were accompanied by a pre-spawning decline in AVT and AVT receptor expression in the brain and testis. Overall, these results support the hypothesis that AVT promotes early spermatogonial proliferation while preventing premature FSH-driven maturation. AVT downregulation coincides with the transition to LH-dominant signaling, which is sufficient to drive the final stages of spermiogenesis, highlighting a key local mechanism controlling male fertility in teleosts.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

The reproductive biology of female Pacific halibut (Hippoglossus stenolepis), a key species in the North Pacific Ocean

Planas, Josep V. (*josep.planas@iphc.int*) Oral Presentation
International Pacific Halibut Commission

Abstract: The Pacific halibut (Hippoglossus stenolepis) is a large migratory demersal flatfish species that occupies a top trophic role in the North Pacific Ocean and Bering Sea ecosystems, where it supports various fisheries. Mature Pacific halibut females migrate from the continental shelf to deep offshore areas of the continental slope for spawning in the winter and return to the continental shelf after spawning. Pacific halibut produces pelagic eggs that are transported by oceanic currents over a period of up to 6 months to shallow nursery areas where post-metamorphic larvae settle and grow to the juvenile stage. In the Northeastern Pacific Ocean, Pacific halibut is currently managed as a single stock by the International Pacific Halibut Commission (IPHC) and reproductive inputs are critical for its annual stock assessment. Over the last few years, the IPHC has investigated in detail a number of important reproductive characteristics of this species, namely qualitative and quantitative aspects of oocyte development in maturing females, the seasonal progression of female maturity throughout the reproductive cycle, the physiological mechanisms responsible for triggering female maturity and ovulation, and the management-relevant relationship between maturity and age in Pacific halibut females, given that age-at-maturity represents a key input for female spawning biomass estimations. In this talk, we will provide a summary of the outcomes from these studies on female Pacific halibut reproductive biology.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

A tale of rainbow trout myostatin – an unconventional journey.

Biga, Peggy[1] (*peggy.biga@txstate.edu*) Oral Presentation
Co-Authors: Addo, Michael[2]; Roberts, Steven[3]; Goetz, Frederick[4]
[1]Texas State University; [2]University of Alabama at Birmingham; [3]University of Washington, United States; [4]NOAA Fisheries

Abstract: Myostatin negatively regulates skeletal muscle growth in vertebrates. In Salmonids, two myostatin transcripts are produced from separate genes resulting in four myostatin isoforms with differential expression in time and space. Early on, Salmonid myostatin genes were referred to as “ovarian” (ov) and “brain/muscle” (b/m) based on Northern and Western blot tissue expression analyses. Later, advances in genetic and genomic technologies allowed for the identification of four distinct myostatin genes, stemming from early genome duplication in bony fish, and a second salmonid-specific duplication about 50-100 million years ago. These myostatin genes were classified and renamed as MSTN-1a, -1b, -2a, and -2b. MSTN-1a, -1b, and -2a are functional paralogs, while MSTN-2b was identified as a pseudogene with two in-frame stop codons. Interestingly, tissue expression analysis of MSTN paralogs has revealed high expression of MSTN-2b in spleen and ovary of female rainbow trout. We therefore have sought to identify if MSTN-2b might be selectively expressed in context specific manners. We show that limiting nutrients can increase MSTN-2b mRNA expression, suggesting there are potential novel functions of MSTN-2b. The fish myostatin journey continues with a new focus on a would-be pseudogene.

REPRODUCTION From reproductive physiology to molecular physiological ecology of fish

From bacterial lipopolysaccharides to behavioural fever in fish; 3 decades of impactful collaborative research with Rick Goetz.

MacKenzie, Simon (*smackenzie70@icloud.com*) Oral Presentation

Abstract: The “fever” response, first described by Hippocrates in 4-5th Century BC, to infection and injury has been maintained throughout at least 600 million years of evolution. Fever, as an epiphenomenon and not a direct cause of disease, can be observed in both ectotherms and endotherms and confers a survival benefit. In mobile ectotherms behavioural fever requires migration to a warmer environment whereas in endotherms significantly increased metabolic rate drives changes in core body temperature. Surprisingly, although fever is a central feature of the immune response, little is currently known of the underpinning mechanisms and how increased temperature influences the activity of the immune response.Here, we will explore how seminal studies, in vitro and in vivo, developed across 3 decades of collaborative research with Rick Goetz and colleagues uncovered different regulatory aspects of the immune response across phylogenetically diverse fish species. From novel pro-inflammatory cytokines to PAMP-driven macrophage activation to novel erythrocyte function to behavioural fever these studies significantly impacted our understanding of the immune response at the molecular, cellular, organ and

whole animal level. Underpinned with the pioneering use of novel molecular technologies combined with classical behavioural and physiological approaches this work ranges from behavioural aspects of individual thermal preference to underlying molecular circuits that build upon the conserved pathways of the immune response to infection.

ADVANCES Recent advances in fish biology and physiology (general session)

Recent advances in fish biology and physiology (general session)

Don MacKinlay

Abstract: Fish biology encompasses a much wider range of topics than what are included in the current list of Congress Symposia. This session is an opportunity for researchers to present their work on topics that do not fit well into that list but are important for the overall understanding of biological mechanisms for fish.

ADVANCES Recent advances in fish biology and physiology (general session)

Get your hands wet! Developing high-impact laboratory exercises for undergraduate fish physiology courses.

Myrick, Christopher (*chris.myrick@colostate.edu*) Oral Presentation
Colorado State University

Abstract: Undergraduate courses in fish physiology can be delivered in a variety of modes, including traditional lecture-based, flipped classrooms, and online or hybrid in-person/online models.Regardless of the mode of instruction, the retention of core principles can be enhanced by the inclusion of laboratory exercises that give students hands-on experience working with fish or simulated fish and taking physiological measurements from them. When considering potential laboratory exercises, instructors need to consider a number of variables, including the availability of specialized research equipment, the availability of suitable species of fish, the local regulatory environment, the amount of time available for setting up and running the laboratory exercise, and the degree of technical aptitude required of participants. In this presentation I will describe a series of laboratory-based exercises that have allowed students to gain experience with fish tagging and measurement, measuring fish growth and diets, fish swimming, fish passage, measuring thermal preference and thermal tolerance limits, exploring fish endothermy, bioenergetic simulations, and basic fish respirometry.I will discuss what made the exercises successful (or unsuccessful), provide suggestions on how some of these exercises can help collect pilot data for future research studies, and cover some of the lessons learned in 32 years of teaching undergraduate fish physiology labs.

ADVANCES Recent advances in fish biology and physiology (general session)

Internal heating rates do not directly reflect water temperature in rainbow trout during CTmax trials

Sutherland, James (*parker39@student.ubc.ca*) Oral Presentation
Co-Author: Bouyoucos, Ian and Brauner, Colin J.
The University of British Columbia

Abstract: Critical thermal maximum (CTmax) is a widely used measure of acute thermal tolerance in fish. Most methodologies to determine CTmax use heating rates between 0.1-0.3ºCmin-1, until fish experience loss of equilibrium (LOE). It is generally assumed that these heating rates are low enough to ensure thermal equilibrium between the internal body temperature of the fish and the surrounding water temperature, but fast enough to prevent thermal acclimation. The former assumption is rarely reported in literature, and recent studies using internal temperature loggers have raised question about CTmax methodology. This study used relatively large (1.5-1.8kg) rainbow trout (Oncorhynchus mykiss) to test this assumption. First, dead fish were implanted with temperature probes in three tissues (brain, dorsal epaxial muscle, and viscera), and real-time temperature was continuously recorded in a standard CTmax protocol using a heating rate of 0.12ºCmin-1. In a second experimental series, live fish were subjected to a similar CTmax experiment until LOE, at which point fish where euthanized and temperature in the same tissues was recorded within two minutes. This allowed a comprehensive assessment of internal tissue heating rates during CTmax, and tested the assumption of thermal equilibrium between the fish and the water. Results indicate that only the brain equilibrates to water temperature using heating rates that are considered low (0.12 ± 0.02ºC), while muscle and viscera temperature where significantly colder than water temperature at CTmax (0.83ºC & 2.1ºC respectively). These data may be important in interpreting CTmax data, as well as its underlying cause in the organism.

ADVANCES Recent advances in fish biology and physiology (general session)

Growth, sexual maturation, and adult body size are linked with the ontogenetic development of aerobic metabolic scope in fish

Norin, Tommy (*tnor@aqua.dtu.dk*) Oral Presentation
Technical University of Denmark

Abstract: Aquatic ectotherms are challenged by global warming and declining oxygen levels, but the physiological determinants of the associated earlier maturation and reduced body size remain unknown. Several frameworks link the body-mass-scaling of metabolic rates with life-history traits, including growth, body size, sexual maturation, and reproduction, but it is debated whether the ontogenetic development of these traits is governed by hard physiological limits, such as oxygen limitation, or adaptive co-optimisation. We quantified ontogenetic growth trajectories, age and size at sexual maturation, and metabolic scaling of 53 individual Trinidadian guppies (Poecilia reticulata)

measured longitudinally from birth to adulthood over 6 months. Sexual maturation could be identified in males, whose growth plateaued out during the study, while female growth continued to accelerate. For both sexes, individual standard (maintenance) metabolic rate (SMR) scaled steeper with body mass than maximum metabolic rate (MMR), resulting in metabolic scope (factorial aerobic scope, FAS=MMR/SMR) declining through ontogeny. Individuals whose FAS decreased the most had lower growth rates and, for males, matured at smaller body sizes and became smaller adults. Males retained high FAS (>5) at maturation and female growth was accelerating, despite being faced with the same reductions in FAS as males, inconsistent with oxygen limitation reducing growth. Overall, our findings link steep declines in metabolic scope to reduced body size at sexual maturation and reduced asymptotic size. They further suggest that covariation between metabolic scaling and life-history is not constrained by hard physiological limits but reflects dynamic shifts in energy allocation that can differ between dimorphic sexes.

ADVANCES Recent advances in fish biology and physiology (general session)

Open-source modular respirometry platform for measuring metabolic rate in fishes: PumpResp, SensResp, and SwimResp

Morozov, Sergey[1] (*sergey.morozov@helsinki.fi*) Oral Presentation
Co-Authors: Prokkola, Jenni[2]; Illing, Björn[3] and Primmer, Craig[1]
[1]University of Helsinki; [2]Natural Resources Institute Finland (LUKE), [3]Thünen Institute of Fisheries Ecology

Abstract: Metabolic physiology is a key determinant of organismal fitness, as metabolic rate constrains the energetic allocation available for growth, locomotion, and reproduction. Whole-body aquatic respirometry, which measures oxygen consumption, remains a standard and robust method for estimating metabolic rate in fishes. However, despite the conceptual simplicity of the technique, commercially available respirometry systems are often prohibitively expensive and rely on proprietary designs, limiting accessibility, modification, and integration with complementary instrumentation required for novel ecophysiological research. Advances in open electronics have facilitated the development of affordable laboratory tools across biological disciplines, yet existing non-proprietary respirometry solutions typically function as extensions to commercial systems or are tailored to narrow experimental contexts. Here, we introduce a universal, well-documented, and highly customizable open-source respirometry platform comprising three hardware modules: PumpResp, SensResp, and SwimResp. Each module is constructed from low-cost generic components and follows a minimalistic design that can be assembled without extensive engineering expertise. PumpResp provides automated control of water pumps for intermittent respirometry systems compatible with optical meters and chambers ranging from 20 mL to 5 L. SensResp functions as an open-source oxygen meter capable of interfacing with low-cost standalone sensors and offers substantial potential for further customization.SwimResp enables precise control of swim-tunnel respirometry, including programmable swimming-speed protocols. All modules underwent experimental validation using Atlantic salmon (Salmo salar) and

rainbow trout (Oncorhynchus mykiss) ranging from 0.5 to 349 g. This open-source platform reduces equipment costs by an order of magnitude, broadens access to respirometry methodologies, and enables various experimental applications, including field deployment and remote monitoring.

ADVANCES Recent advances in fish biology and physiology (general session)

Effects of warm exposure on swimming performance and aerobic metabolic capacity in juvenile Yukon River Chinook salmon

Snyder, Augustus (*amsnyder7@alaska.edu*) Oral Presentation
Co-Authors: Wagner-Jones, Amelia; Barreiros Scatamburlo, Larissa and Gilbert, Matthew
University of Alaska Fairbanks

Abstract: Rapid warming of Arctic and sub-Arctic freshwater ecosystems is increasing the frequency and severity of thermal stress experienced by juvenile Pacific salmon during freshwater rearing. For Yukon River Chinook salmon (Oncorhynchus tshawytscha), elevated temperatures threaten aerobic metabolic capacity and swimming performance, key determinants of growth, predator avoidance, and survival. These effects and the mechanistic links between temperature acclimation and performance remain poorly resolved for northern populations. To address this gap, we quantified the effects of prolonged warm temperature exposure on swimming performance, resting and maximum aerobic performance, and aerobic capacity (i.e., aerobic scope) in juvenile Yukon River Chinook salmon. Rearing temperatures spanned ecologically relevant summer temperatures from 12.5 to 22.5 °C. Preliminary results suggest thermal acclimation significantly altered both swimming performance and metabolic capacity; I will be presenting the emerging results of this study. By linking swimming performance and aerobic metabolism across realistic thermal exposures, this study provides a mechanistic framework for understanding how warming freshwater habitats may limit juvenile Chinook salmon performance in northern river systems. These findings have implications for forecasting population vulnerability under climate change and for informing conservation and management strategies for culturally and ecologically important Yukon River Chinook salmon stocks.

ADVANCES Recent advances in fish biology and physiology (general session)

Changes to group dynamics reflect changes in individual behaviour under thermal stress in a model fish species

Tiddy, Izzy C.[1] (*isabelle.tiddy@glasgow.ac.uk*) Oral Presentation
Co-Authors: Ioannou, Christos C.[2]; Torney, Colin[1]; Webster, Michael M.[3] and Killen, Shaun[1]
[1]University of Glasgow, Glasgow, [2]University of Bristol, [3]University of St Andrews

Abstract: As global temperatures rise, the ways in which organisms respond and the wider implications of these responses at the level of social groups are of increasing interest. While temperature can affect individual and group level social behaviour in fish species,

the links between individual and group level behaviour under thermal stress remain poorly understood. Elucidating these links is key for understanding how individual traits translate to interactions with the wider ecosystem. Here, we investigated whether effects of temperature on individual-level behaviour would be reflected in changes in group dynamics in rummy-nose tetras, Hemigrammus rhodostomus, using tracking of fish with quantified behavioural traits. We further tested whether measures of individual exploratory behaviour, sociability, and activity level would impact position or interactions of these same individuals within groups at three temperatures: 24°C, 27°C, and 30°C. Less-social, less-active individuals that displayed greater exploratory behaviour were more likely to swim at the front of the shoal, possibly giving these individuals greater influence on group decision making. Speed and polarisation at the group and individual levels were increased at higher temperatures, however this trend was reversed in social, less-active fish, and larger fish in the case of group polarisation. Our results indicate that behavioural traits and size of individuals inform the role of those individuals within groups and group-level behaviours, but that these relationships are also dependent on environmental factors. Our findings highlight the importance of incorporating both the social and abiotic environment to studies of fish behaviour.

ADVANCES Recent advances in fish biology and physiology (general session)

Does the rate of temperature change affect thermal plasticity in fishes?

Welsh, Emily (*emily.welsh@unb.ca*) Oral Presentation
Co-Author: Hartenstein, Peyton; Speers-Roesch, Ben
University of New Brunswick Saint John

Abstract: To cope with daily or seasonal temperature challenges, fish can adjust their physiological rates and thermal tolerance through thermal acclimation. Few studies have considered how thermal plasticity manifests under gradual temperature change, which better mimics natural seasonal change versus the traditional rapid-stable approach to studying thermal acclimation. We investigated whether the rate of temperature change affected thermal plasticity during simulated winter cooling and summer warming in mummichog (Fundulus heteroclitus) and brook char (Salvelinus fontinalis). Cooling fish were acclimated to either: i) rapid-stable cold exposure from 14°C to 3°C at ~6°C/day (in 2 days) and then held stable at 3°C for 56 days, or ii) gradual cold exposure from 14°C to 3°C at an average seasonally realistic rate of 0.2°C/day to reach 3°C after 56 days. Warming fish were exposed identically except with warming to either 20°C or 26°C for brook char and mummichog, respectively. Standard metabolic rate (SMR), maximum metabolic rate, and critical thermal maximum (CTmax) of fish were measured at comparable timepoints over 3 months. For both species during 3°C exposures, the metabolic rates and CTmax of fish exposed to the gradual treatment converged with the rapid-stable fish. However, mummichog showed no evidence of metabolic thermal compensation, whereas the SMR of brook char displayed a delayed thermal compensation. The warming experiments are on-going. Our results thus far suggest that the rate of temperature change does not

influence the final phenotype resulting from thermal acclimation in fishes, but may influence its temporal manifestation.

ADVANCES Recent advances in fish biology and physiology (general session)

Modeling 125 years of spatial and temporal variability in climate and fish assemblages of temperate inland lakes

Lucas, Kelsey[1] (*kelsey.lucas@ucalgary.ca*) Oral Presentation
Co-Author: Breck, James[2]; Alofs, Karen[3]
[1]University of Calgary; [2]Michigan Department of Natural Resources, [3]University of Michigan

Abstract: Understanding the level of spatial and temporal variation in lake temperature and, by proxy, fish species assemblages is imperative for anticipating responses to environmental change. However, the degree of temperature variation that we can expect among temperate inland lakes and hence the interannual variation in fish assemblages is poorly understood. Here, we take a modeling approach that leverages publicly available, historical climate observations to hindcast the annual thermal regime of inland lakes over a 125-year period and assess water temperature variability across space and time. We use Michigan – encompassing 6394 inland lakes that are situated across the <10 deg C temperature band separating cold, cool, and warm guilds – as a model system. We then use our hindcasted thermal regimes and Michigan's ecological lake classification scheme to infer the lake fish assemblages that have been favored over space and time. We find a subtle but significant long-term warming trend that appears to be driven by changes in seasonality. We also find substantial short-term variation in lake temperature that has increased over the last several decades. We suggest that, as a result, cold and cool lake fish assemblages have become less persistent in temperate inland lakes. Our results can be used to identify how responsive individual lakes are to climate variability as well as the assemblages that are expected to be favored under future climate change. Such information is essential for developing effective adaptation strategies.

ADVANCES Recent advances in fish biology and physiology (general session)

Application of environmental RNA in the ecophysiological studies of fish

Wong, Marty Kwok-Shing (*martywong@aori.u-tokyo.ac.jp*) Oral Presentation
Co-Authors: Aminaka, Yuto and Hyodo, Susumu
The University of Tokyo

Abstract: The use of environmental DNA (eDNA) in monitoring wildlife diversity and community structures has been widely accepted as a cost-effective, robust, and non-invasive method in government bodies and academics. Fish leave their eDNA in the water via epithelial shredding, mucus secretion, excretion, gamete release, etc. The key success of eDNA is that DNA from different cell types is identical, which can be used as a unique fingerprint to identify the host fish. Although eDNA can be used identify the fish species, the fish activity or physiological status could not be deduced.Environmental RNA (eRNA),

however, has the potential to infer the status and shed lights on the activity of the fish species because mRNA is functional- and tissue-specific. We examined gonad-specific genes in medaka with reference to literature and transcriptome databases and developed quantitative real time PCR assays to measure their transcript levels in the water. The eRNA level ofklhl10, a transcription factor that is specific in sperm development, was found to be abundant when courtship and mating behavior was observed, suggesting that the release of semen has increased theklhl10level in the water. With the tissue specificity ofklhl10and the coincided timing of detection in the water, the presence ofklhl10can be an eRNA marker for spawning event in fish. In the future, we aim to explore other use of eRNA such as deducing stress condition of fish species with appropriate eRNA markers, which may benefit fish farming and disease control.

ADVANCES Recent advances in fish biology and physiology (general session)

Impacts of body size and parasite load on the preferred temperature and avoidance temperatures of the diadromous fish Galaxias maculatus

Gradito, Maryane (*m.gradito@deakin.edu.au*) Oral Presentation
Co-Authors: Clark, Timothy
Deakin University

Abstract: Aquatic ectotherms often experience wide thermal variation in their habitats, and motile species such as fishes can behaviourally select temperatures that optimise physiological performance in a context-dependent manner. However, quantifying drivers of thermal selection of fishes can be challenging in the natural environment, particularly for small-bodied species. In laboratory settings, two-chamber shuttlebox systems are commonly used to quantify behavioural thermoregulation, preferred temperature (Tpref), and upper and lower avoidance temperatures (Tavoid). Here, we sought to understand important factors influencing thermal selection in the ecologically important Australian fish Galaxias maculatus using a shuttlebox. Specifically, we tested whether measures of thermal selection were influenced by (1) fish size and metabolic scaling, (2) fish activity levels, and (3) shuttlebox starting temperatures. Additionally, we examined whether natural parasite loads (nematode; Eustrongylides sp.) influenced thermal selection and activity levels, predicting that heavier infections might alter movement behaviour or induce behavioural fever or chill. We found a Tpref range of 13.54–21.68 °C (mean 17.25 °C), a lower Tavoid range of 5.78–16.64 °C, and an upper Tavoid range of 19.57–28.14 °C, indicating substantial intraspecific variation in thermal selection. Larger fish tended to select lower preferred and upper avoidance temperatures, consistent with potential metabolic scaling effects. Activity level and starting temperatures did not influence Tpref or Tavoid, demonstrating the robustness of the shuttlebox protocols. We did not find any impact of natural parasite load on the thermal preference of G. maculatus. Overall, our study refines shuttlebox approaches and provides high-resolution insights into drivers of thermal selection in eurythermal fishes.

ADVANCES Recent advances in fish biology and physiology (general session)

The influence of food availability on metabolic thermal plasticity in overwintering brook char (Salvelinus fontinalis)

Hartenstein, Peyton[1] (*peytonhartenstein1@gmail.com*) Oral Presentation
Co-Authors: Welsh, Emily[1]; Speers-Roesch, Ben[1] and Gilbert, Matthew[2]
[1]University of New Brunswick, [2]University of Alaska Fairbanks

Abstract: Food availability can alter the metabolism of fishes and is emerging as an important modulator of thermal plasticity. For example, starvation commonly results in metabolic rate depression while our recent work on brook char (Salvelinus fontinalis) showed that fish exposed to starvation and cold, common co-occurring stressors over winter, lacked the thermal compensation of metabolic rate that is common in this species and other winter-active fishes when they can feed. Interestingly, winter-inactive species, which voluntarily feed little over winter (unlike winter-active species), also lack metabolic thermal compensation. We hypothesize that masking of thermal compensation by food deprivation, whether involuntary or voluntary, is universal among overwintering fishes and results from the counteracting effects of starvation and cold compensation on metabolic rate. Previous studies were unable to directly test this hypothesis because starvation and cold exposure were applied simultaneously, which is ecologically-relevant but complicates identification of each factor's effect. Here, using winter-active brook char, we sought to resolve this previous limitation by measuring standard metabolic rate (SMR) repeatedly within individual fish that were either fed daily or starved during sequential exposure to 14°C and then 2°C. SMR was measured at 14°C before starvation began and 3 weeks later to isolate the starvation effect, and then at 2°C following acute cooling and 6 weeks later to identify whether cold compensation occurred even in starved fish. Our work provides clearer insight into how food scarcity and cold interact to shape metabolic plasticity in fish under winter conditions.

ADVANCES Recent advances in fish biology and physiology (general session)

The impact of environmental conditions on Atlantic sturgeon (Acipenser oxyrinchus oxyrinchus) habitat use

Gonzalez, Kelly[1] (*kairvin@vims.edu*) Oral Presentation
Co-Authors: McDowell, Jan[1]; Adkins, Dana[2]; Hunt, Joe[2] and Dichiera, Angelina[1]
[1]Virginia Institute of Marine Science, [2]Chickahominy Tribe

Abstract: Atlantic sturgeon (Acipenser oxyrinchus oxyrinchus) are endangered across their native range, and a major challenge to repopulation efforts is the lack of suitable habitat in natal rivers. A distinct population segment (DPS) of Atlantic sturgeon in Chesapeake Bay utilizes at least four of its tributaries for spawning migrations, including the James River. Suitable habitat is important not only for spawning but also for providing resources for early life stage success. Environmental changes such as warming and reduced oxygen availability are physiologically costly and can have cascading effects on migration, reproduction, and abundance. Our understanding of Chesapeake Bay DPS habitat use is

limited by high effort monitoring strategies and lack of habitat information for upper tributaries. To address these challenges, this study aims to characterize Atlantic sturgeon's presence and habitat use in the Chickahominy River, an offshoot of the James River, using environmental DNA (eDNA). eDNA provides a lower effort, highly sensitive alternative to traditional monitoring strategies. Water samples and habitat characteristics are collected monthly from the Chickahominy and James Rivers. Quantitative PCR will be used to quantify relative abundance of Atlantic sturgeon cytochrome b gene and generalized linear models will be used to determine if environmental conditions (temperature, dissolved oxygen, and salinity) and/or time of year are predictors of eDNA abundance. Atlantic sturgeon eDNA is expected to be found in the Chickahominy River, with higher abundance in spring and fall, indicating spawning aggregations. Further, characterizing how environmental conditions impact Atlantic sturgeon abundance will provide critical information on habitat needs.

ADVANCES Recent advances in fish biology and physiology (general session)

Oxygen uptake of rainbow trout during acute warming: effects of oxygen supersaturation and aerobic exercise

Bellinger, Imogen (*imogen.bellinger@gmail.com*) Oral Presentation
Co-Author: Raby, Graham; Bauer Reid, Heather
Trent University

Abstract: Naturally occurring oxygen supersaturation (hyperoxia) could provide refuge for fish from heat waves if thermal tolerance is limited by the ability to uptake sufficient oxygen to meet cellular oxygen demands during acute heat stress, as previously proposed. While several studies have assessed warming tolerance (CTmax) in water breathers under hyperoxia, none have assessed whether CTSmax (warming tolerance during aerobic exercise) is affected by hyperoxia. Here, we assessed whether hyperoxia could increase the thermal tolerance and metabolic rate of rainbow trout (Oncorhynchus mykiss) both at high swimming speeds (CTSmax; 85% of maximum swimming speed) and in traditional CTmax trials. To do so, n=16 rainbow trout were subjected to thermal tolerance trials under two treatments: hyperoxia (150% dissolved oxygen [DO]) and normoxia (100% DO). Under these treatments, fish were warmed at a rate of 2°C hour-1, with estimates of oxygen consumption ($\dot{M}O_2$) generated for each 30 min (1°C) increment. Hyperoxia increased thermal tolerance in CTSmax trials by ca. 0.6°C but had no effect on CTmax. Surprisingly, we found no effect of hyperoxia on $\dot{M}O_2$ during the CTSmax trials, suggesting an increase in oxygen uptake was not responsible for the increase in warming tolerance under hyperoxia. Interestingly, although all fish increased their $\dot{M}O_2$ during the acute warming trials, many did not reach $\dot{M}O_{2,max}$ based on earlier critical swimming speed (Ucrit) trials at a benign temperature. Together, our results provide insight into the effects of ecologically relevant levels of oxygen saturation on thermal tolerance in fish.

ADVANCES Recent advances in fish biology and physiology (general session)

Effects of experimental infections on host metabolism and oxidative damage in naïve and experienced populations of pumpkinseed sunfish (Lepomis gibbosus)

Mélançon, Vincent (*vincent.melancon@umontreal.ca*) Oral Presentation
Co-Author: Binning, Sandra; Fortin-Hamel,Liana and Breton, Sophie
Université de Montréal

Abstract: Environmental differences can drive population-level divergence in a number of physiological traits. Exposure to parasites, in particular, can impact host energy allocation, which may result in altered metabolic activities among populations experiencing different levels of infection risk. The degree to which parasite exposure and infection drive population-level differences in whole organisms and sub-cellular metabolic rates has rarely been explored. Using field mesocosms (cages) in natural lakes, we investigated cellular mechanisms underlying changes in oxygen uptake following parasite infection in a parasite-naïve and a parasite-experienced population of pumpkinseed sunfish. Oxygen uptake in fish from both populations was measured in respirometers before and after spending six weeks in cages placed in lakes with either a naturally high or negligible exposure to cestode and trematode infections. Whole-organism estimates of metabolism were then linked to cellular responses including oxidative damage, maximal mitochondrial enzyme activity, and antioxidant activity. Susceptibility to infection differed between populations, with the highest experimentally induced parasite infections occurring in the naïve population. We also found differences in both whole-organism and sub-cellular estimates of aerobic metabolism between populations depending on infection treatment. Population-level differences in infection susceptibility may reflect metabolic adaptations in parasite experienced populations that reduce the costs of infection, resulting in lower oxidative damage and smaller increases in mitochondrial activity. Given the predicted increased in parasite virulence/intensity due to climate change, our results highlight the importance of understanding population-level variation in physiological performance measures across levels of biological organization to better understand infection dynamics in a changing world.

ADVANCES Recent advances in fish biology and physiology (general session)

Abiotic and biotic drivers of declining flounder abundance, distribution, and condition

Zavell, Max[2] (*mzavell@umassd.edu*) Oral Presentation
Co-Author: Schultz, Eric[1]; Helmer, Katherine[1]; Mosca, Kelli[3]; Gates, Matthew[3]; Gottschall, Kurt[3]; Batta-Lona, Paola[1]; Baumann, Hannes[1] and Klarian, Sebastián[4]
[1]University of Connecticut; [2]University of Massachusetts Dartmouth, [3]Conneticut Department of Energy and Environmental Protection, [4]Universidad Andres Bello

Abstract: Concurrent anthropogenic changes have restructured marine community assemblages and trophic dynamics in Long Island Sound (LIS, between Connecticut and New York, USA). Among the affected taxa are species of flatfishes such as Winter Flounder

(Pseudopleuronectes americanus) and Summer Flounder (Paralichthys dentatus). Winter Flounder populations have collapsed throughout southern New England, and although adult populations of Summer Flounder have remained stable, their recruitment has recently declined. Both species have been designated as “species of greatest conservation need”. To quantify potential drivers affecting Winter and Summer Flounder population dynamics we analyzed over forty years of Long Island Sound Trawl Survey (LISTS) data to assess changes in environmental variables (substrate, temperature, depth, salinity, dissolved oxygen) and species composition (competitors and prey) in LIS and how these changes might affect population dynamics of these species. Further, we developed predictive models that will inform management responses for flatfish species under continued abiotic and biotic change. Between 1990 and 2024, mean Winter Flounder abundance decreased by ~97%, while mean Summer Flounder abundance decreased by ~69% and remained stable through 2024. Mean competitor species abundance increased by ~295% while prey species abundance decreased by ~74% between 1995 and 2024. Initial analysis suggests that both species preferentially occupy habitats at depths < 20 meters. Declines in the abundance of both species are associated with warmer bottom water temperatures and increases in competitor abundance. Abiotic and biotic change will have an ongoing effect on flatfish habitat usage and abundance.

ADVANCES Recent advances in fish biology and physiology (general session)

Natural microalgal metabolites promote recovery of insulin-producing β-cells in zebrafish.

Ferreira-Martins, Diogo (*dmartins@ciimar.up.pt*) Poster Presentation
Co-Authors: Silva, Maria; Moxo, Javier and Urbatzka, Ralph
University of Porto

Abstract: Diabetes is a metabolic disease that affects hundreds of millions of people worldwide and contributes substantially to global morbidity and mortality. Addressing diabetes requires comprehensive global health strategies which include finding alternative solutions for treatment and prevention. Microalgae generate numerous metabolites with potential to be exploit for biotechnological and biomedical applications. In this work, we analyzed organic extracts of 11 microalgae species for diabetes therapeutics. Dichloromethane/methanol (DCM/MeOH) and acetone extracts were prepared from microalgae. Transgenic zebrafish [Tg(ins:GFP-T2A-NTR3, cryaa:mCherry)] were exposed to the extracts (25 µg/mL) for 24 hours to assess the recovery of insulin-producing β-cells following metronidazole (MTZ) induced ablation and nitroreductase expression in β-cells. DCM/MeOH extracts from Scenedesmus obliquus, Arthrospira platensis, Nannochloropsis oceanica, Nanofrustulum shiloi, Phaeodactylum sp., and Chlorococcum sp. increased the number of pancreatic β-cells compared to the MTZ treated group. LC-MS/MS was used to determine the chemical composition of the extracts followed by in silico molecular networking through the Global Natural Product Social Molecular Networking (GNPS) platform to identify potential bioactive compound families. To investigate the mechanisms of action of the active extracts, genes involved in glucose regulation were analyzed. The

identified compounds may support development of novel nutraceuticals for improved human health. This work was financially supported by “Pacto da Bioeconomia azul” (Project No. C644915664-00000026) within the WP5 Algae Vertical, funded by Next Generation EU European Fund and the Portuguese Recovery and Resilience Plan (PRR), under the scope of the incentive line “Agendas for Business Innovation” through the funding scheme C5 - Capitalization and Business Innovation.

ADVANCES Recent advances in fish biology and physiology (general session)

Evidence for subfunctionalization of excitation-contraction coupling partners following the salmonid-specific genome duplication event

Franck, Jens[1] (*j.franck@uwinnipeg.ca*) Poster Presentation
Co-Author: Sadaya, Paula[1]; Robinson, Sean[2]; Good, Sara[1]
[1]University of Winnipeg; [2]University of Guelph

Abstract: Excitation-contraction (EC) coupling relies on the activation of the sarcoplasmic reticulum (SR) calcium release channel or ryanodine receptor (RyR1). In vertebrate skeletal muscle the RyR is mechanically gated by the voltage-gated L-type calcium channel (CaV1.1) in the sarcolemmal membrane in a mechanism termed depolarization-induced calcium release (DICR). The teleost-specific whole genome duplication (ts-WGD) generated duplicated genes (paralogs) of both the ryr1 calcium release channel and the main a1S subunit of the CaV1.1 voltage sensor. In all teleost species examined to date, ryr1a and a1S-a are expressed in slow-twitch red muscle, while the ryr1b and a1S-b paralogs are expressed in fast-twitch white muscle, an example of subfunctionalization following a gene duplication event. The Salmonidae family has undergone an additional genome duplication event approximately 100 mya, resulting in the retention of two ryr1 paralogs, ryr1a-1 and ryr1a-2 as well as two a1S paralogs, a1S-1 and a1S-2. We investigated the expression of the full complement of ryr and a1S paralogs in slow and fast twitch muscle tissues of rainbow trout (Oncorhynchus mykiss) using quantitative PCR (qPCR) of relative transcript levels as a proxy for gene expression. In addition to tissue-specific expression we examined temperature effects on transcript levels. The additional ryr1a and a1S paralogs found in trout reveal expression patterns that are both consistent and inconsistent with those of other teleost fish. We discuss the inconsistencies and their functional significance to EC coupling in salmonids.

ADVANCES Recent advances in fish biology and physiology (general session)

The feasibility of acoustic telemetry in longfin smelt, Spirinchus thaleichthys, using miniaturized acoustic transmitters

Gonzales, Sebastian (*sggonzales@ucdavis.edu*) Poster Presentation
Co-Author: Fangue, Nann[1]; Steel, Anna[1]; Cocherell, Dennis[1]; Deng, Zhiqun (Daniel)[2]; Wilder, Rick[3]; Schreier, Brian[4] and Lewis, Levi[1]
[1]University of California Davis, [2]Pacific Northwest National Laboratory, [3]ICF International, [4]California Department of Water Resources

Abstract: Longfin smelt are a pelagic, small-bodied forage fish distributed along the Pacific coast of North America. The San Francisco Estuary of California supports the Bay-Delta Distinct Population Segment of longfin smelt, a genetically distinct source population and the southernmost reproductive population of the species. Once among the most abundant fishes in the San Francisco Estuary, long-term monitoring programs have documented a precipitous decline in Bay-Delta longfin smelt abundance since the 1960s, with current estimates at less than one percent of pre-1980 levels. Recent advances in transmitter miniaturization by the Pacific Northwest National Laboratory, including the development of the Shad Tag (0.05 g), have created new opportunities to apply acoustic telemetry to the study and conservation of wild longfin smelt populations. Prior to implementing acoustic telemetry as a conservation tool for longfin smelt, the effects of acoustic tagging surgery and tag-carrying burden must be evaluated under controlled conditions to verify that tagged individuals are not behaviorally or physiologically inhibited and that telemetry-derived data are representative of the broader, untagged population. Here, we present results examining the effects of acoustic tag implantation on survival and swimming performance (measured as critical swimming speed, Ucrit) in wild-caught Bay-Delta longfin smelt, using tagging methods adapted from those developed for the sympatric Delta smelt. These findings inform assessments of the feasibility of acoustic telemetry as a research and conservation tool for longfin smelt.

ADVANCES Recent advances in fish biology and physiology (general session)

Distributional responses of European sea bass to operational floating wind turbine noise in a semi-natural outdoor mesocosm system.

Lefrancois, Christel[1] (*christel.lefrancois@univ-lr.fr*) Poster Presentation
Co-Author: Millot, Morgane[1]; Helloco, Loïc[2]; Dubillot, Emanuel[1], Artero, Celine[3,4]; Bellier, Benjamin[1]; Carras, Karla[1]; Boucard, Johann[1] and Le Bourhis, Enored[5]
[1]La Rochelle University, [2]CREOCEAN, [3]BOREA-MNHN, [4]HEPIA Suisse, [5]Fondation Open-C

Abstract: Among the various forms of anthropogenic pollution, underwater noise has only recently been recognized as a significant environmental stressor, particularly in the context of the rapid expansion of offshore wind energy developments. Environmental risk assessments are predominantly based on the determination of acoustic thresholds inducing physiological injury, such as temporary and permanent threshold shift. However,

biological effects are expected at sound levels below those inducing measurable auditory damage, such as behavioral alterations that may ultimately affect individual fitness.
The present study aimed to investigate the effects of sub-injury sound levels, with a particular focus on behavioral disturbances reflected by changes in spatial distribution. We examined distributional responses in groups of European sea bass exposed to in-situ recordings representativeof floating offshore wind turbine noise.
The study was conducted under semi-natural conditions using outdoor experimental tanks (260 m^3 each; water depth: 1.30 m). Ten adult individuals equipped with passive tags were allowed to move freely between two adjacent tanks connected by a 1 m-wide channel fitted with two telemetry antennas. Acoustic playback was played in one of the tanks, thereby creating an exposed area and a refuge area separated by the channel. Fish spatial distribution was monitored during three successive phases : (1) an acclimation phase without noise emission, (2) exposure to ambient sea noise, and (3) exposure to wind turbine noise superimposed on ambient sea noise.
Preliminary results indicate that noise exposure can significantly alter fish spatial distribution, with a more pronounced avoidance response observed under wind turbine noise conditions.

ADVANCES Recent advances in fish biology and physiology (general session)

Reproductive steroid patterns during late-stage oocyte maturation in wild female burbot (Lota lota)

Leonard, Jill[1] (*jileonar@nmu.edu*) Poster Presentation
Co-Author: Legacki, Erin[2]; Anderson, Lars[1]; Hill, Anna[1]
[1]Northern Michigan University; [2]USDA

Abstract: Burbot, the only freshwater gadiform, retains many marine-like reproductive habits, including spawning in cold temperatures with a very short reproductive window. This short spawning window presents vulnerabilities to the species because it requires precise coordination of migration and reproduction, which is completed in a single spawning event. We know little about the physiological mechanisms for this tightly controlled strategy, including its hormonal control. Wild burbot females were captured during their upstream migration in the Chocolay River in hoop nets set overnight. Fish were measured on the riverbank and then blood and oocytes were collected. Fish were then returned to our laboratory and maintained at 2oC with subsequent blood and oocyte sampling daily until they spawned (either naturally or artificially). Oocytes were characterized to developmental stage. Blood and oocytes were frozen at -70C until they were assessed using a panel for twenty reproductive steroids using mass spectrometry. In plasma, THP, 1 deoxycorticosterone, 11 deoxycortisol, corticosterone, and testosterone showed declines as oocyte stage approached maturation while plasma cortisol and cortisone increased. These patterns persisted in both immediately caught fish and in fish held in the lab for maturation. In oocytes, steroid patterns were similar to plasma for the above hormones, but progesterone, pregnenolone, 12,20 dihydroprogesterone, and 17 hydroxyallopregnenolone also declined during maturation. While our data are preliminary,

they demonstrate substantial steroidal changes in both the plasma and oocytes in the several days prior to spawning, with significant differences apparent between plasma and developing eggs.

ADVANCES Recent advances in fish biology and physiology (general session)

Tracking American Eel habitat use using non-lethal stable isotope analysis

Simon, Sage[1] (*meadowsimon@live.ca*) Poster Presentation
Co-Author: Sacobie, Charles[1]; Roloson, Scott[2]
[1]University of New Brunswick; [2]Department of Fisheries and Oceans Canada

Abstract: Recent research has revealed that American eels (Anguilla rostrata) are a facultative catadromous species, as individuals may reside in or move among freshwater, estuarine, and coastal habitats throughout their lives. However, the patterns driving these habitat shifts remain poorly understood. Combined with the species' panmictic population structure, this uncertainty makes it difficult to obtain accurate population estimates and develop effective conservation strategies. Stable isotope analysis (SIA) has become a valuable tool for studying fish migrations across salinity gradients. Carbon isotope ratios ($\delta^{13}C$) are reflected from consumed prey items and often follow predictable patterns across freshwater to marine environments, while nitrogen isotope ratios ($\delta^{15}N$) provide information about trophic position within the food web. When multiple tissues with different turnover rates are analyzed, SIA can reveal habitat use across different time frames. As uncertainty surrounding the status of American eel populations persists, non-lethal methods for studying their ecology are increasingly important. The objective of this study is to determine patterns of habitat use and habitat switching in American eels using a non-lethal SIA approach. Blood, fin, and muscle tissues were collected from eels captured in two river systems in Prince Edward Island to examine their short- and long-term habitat use and patterns, and $\delta^{13}C$ and $\delta^{15}N$ from these tissues will be compared with the isotopic signatures of the habitats to assess their habitat use. This research will provide new insight into eel habitat use and movement patterns, supporting improved conservation and management of this ecologically and culturally significant species.

ADVANCES Recent advances in fish biology and physiology (general session)

Single parental cold exposure drives transgenerational rewiring of energy allocation across the tissue network in tilapia

Wang, Min-Chen[1] (*mcwinlab@gmail.com*) Poster Presentation
Co-Author: Tseng, Yung-Che[2]
[1]Christian-Albrechts-University Kiel; [2]Academia Sinica

Abstract: Organisms respond to environmental change through complex and coordinated tissue networks. They dynamically allocate limited energy fuel among tissues to support necessary physiological adjustments. The present study investigated the transgenerational effect on dynamic energy allocation across tissues in tilapia (Oreochromis mossambicus).

The transcriptomic profiling shows that a single lifetime cold exposure in the parental generation was sufficient to induce pronounced transcriptional reprogramming in their offspring. Under normothermic conditions, offspring exhibited overwintering-like molecular signatures, indicating an anticipatory physiological state. This highlights the sensitivity of integrated physiological networks to episodic stressors. Consequently, it raises important questions about the adaptive significance and potential costs of anticipatory transgenerational programming.

RESPIRATION Respiratory gas exchange and circulation: The legacy of Dave Randall

Respiratory gas exchange and circulation: The legacy of Dave Randall

Danielle McDonald and Alex Zimmer

Abstract: With five classic papers in 1967 that measured blood pressure, blood gases, and gas exchange in chronically cannulated trout during exercise and hypoxia, Dave Randall, together with Don Stevens and George Holeton, established the modern era of respiratory and circulatory physiology of fish. Building on this legacy, this symposium will highlight the latest advancements in understanding how fish breathe, circulate blood, and adapt to changing environmental conditions (e.g., hypoxia, temperature, salinity). The research presented will incorporate cutting-edge imaging, molecular techniques, and integrative approaches that link gill function, oxygen transport, and environmental adaptation. As a forward-looking discussion of where the field is headed, this symposium will serve to celebrate the visionary scientist that started it all.

RESPIRATION Respiratory gas exchange and circulation: The legacy of Dave Randall

Notes on my travels with Dave

Iwama, George (*georgeiwama@gmail.com*) Oral Presentation

Abstract: My life changed when I met Dave Randall in 1975. This talk will touch on some aspects of my personal and professional life since that day. Dave Randall was a not only my mentor and teacher, but also a personal friend. With apologies to those who have attended the tributes to Dave Randall over the last couple of years, I shall provide a summary of the kind of research the good people in my lab at UBC and I conducted. Interspersed among those research highlights will be glimpses into the personal moments that Dave and I shared. Many of these involved members of our families, but personal and the good people in our labs. The science I will share will be mostly about the stress responses in fishes from around the world. Many in the audience may recognize much younger versions of themselves, or of their mentors and supervisors. Thank you Dave, for everything. Thank you to Dal and Val for organizing this well-deserved tribute to our friend and mentor.

RESPIRATION Respiratory gas exchange and circulation: The legacy of Dave Randall

Insights from 'omics about anoxic crucian carp

Lefevre, Sjannie (*s.l.nilsson@ibv.uio.no*) Oral Presentation
University of Oslo

Abstract: The crucian carp (Carassius carassius) is extremely tolerant to the absence of oxygen. Its metabolically most active organs, such as the brain, can maintain function over extended periods of anoxia. Our research seeks to answer the basic question: what are the cellular and molecular mechanisms involved in regulating the response of the crucian carp to anoxia? To investigate this we take a discovery-driven approach studying the effect of anoxia and reoxygenation on for example the methylome, transcriptome, and translatome, having done most work so far on the brain. Sequencing of both mRNA and ribosome-protected mRNA fragments has, for example, revealed that translation efficiency for some genes changes more rapidly than transcription in response to anoxia and particularly re-oxygenation, giving the crucian carp finer temporal control of the protein pool and hence cellular activities. The data also indicate that increases in mRNA abundance may be buffered by reduced translation efficiency during anoxia, such that overall translation can quickly increase in reoxygenation. Overall, the crucian carp brain shows a large transcriptional response to anoxia and reoxygenation, but given that the brain is a complex and heterogenous tissue, we have also used spatial transcriptomics to explore if the transcriptional anoxia responses differ between brain regions. Based on these mRNA abundance data, we successfully identified brain regions with distinct gene expression patterns and responses to anoxia across the telencephalon, optic tectum, and thalamus, confirming the complexity and heterogeneity of the brain.

RESPIRATION Respiratory gas exchange and circulation: The legacy of Dave Randall

Comparative red blood cell physiology: Uncovering novel cellular mechanisms to understand the impact of climate change on fishes

Harter, Till (*tharter@geomar.de*) Oral Presentation
GEOMAR Helmholtz Centre for Ocean Research Kiel

Abstract: Dave Randall was an "ideas guy". He understood early-on that red blood cells (RBC) are not simply bags full of haemoglobin, but active cells that interact with the vasculature and the tissues. He pioneered the idea that in teleost fishes with highly pH-sensitive haemoglobins, acid-base disequilibrium states in the blood can enhance cardiovascular O2 transport, if carbonic anhydrase is present in the arterial vasculature. More recent work has shown that: i) the RBCs of lower vertebrates, including fishes, have nucleated RBCs that remain transcriptionally active, enabling plastic phenotypic responses to environmental and metabolic stressors; ii) RBCs can regulate their intracellular microenvironment to modulate the function of proteins involved in O2 and CO2 transport; iii) RBCs can sense the conditions of O2 and pH in the microcirculation; and iv) release signaling molecules in response, that increase blood flow through vasodilation, and reduce respiration by inhibiting mitochondrial function. These findings

have led to a novel framework that places the RBC at the centre of an integrative mechanism that can balance microvascular gas transport and metabolic demand. My lab investigates how phenotypic plasticity in RBC function contributes to the response of fishes to acute environmental stressors, and how adaptations at the RBC level may help some populations overcome the chronic threat of climate change. The discovery of novel cellular pathways in vertebrate RBCs will help us understand, predict and mitigate the impacts of climate change on aquatic ecosystems and may find application in the treatment of human conditions related to tissue O2 deprivation.

RESPIRATION Respiratory gas exchange and circulation: The legacy of Dave Randall

Trading athleticism for hypoxia tolerance: Ontogenetic reorganisation of oxygen transport in coral reef fishes

Rummer, Jodie[1] (*jodie.rummer@jcu.edu.au*) Oral Presentation
Co-Author: Downie, Adam[2]; Prescott, Leteisha[3]
James Cook University; [2]Queensland University of Technology; [3]University of Tasmania

Abstract: The assembly of cardio-respiratory systems during early development sets the framework for aerobic performance in fishes; yet, the functional role of these systems during embryogenesis and larval transitions remains poorly understood in tropical species. Using Indo-Pacific coral reef fishes (e.g., Acanthochromis polyacanthus, Amphripion melanopus, Pomacentrus amboinensis), we examined how oxygen transport capacity is constructed and subsequently reprogrammed across early ontogeny. Embryos establish a functional heartbeat within two days of fertilisation, with red blood cell circulation and complex gill structures forming prior to hatching. However, despite early haemoglobin production, experimental haemoglobin disruption revealed no effect on oxygen uptake until after hatching, indicating that, along with immunohistochemistry, pre-hatch gill development primarily supports ionoregulation rather than branchial respiration. Thus, the respiratory system is assembled in advance of functional dependence on haemoglobin-mediated oxygen transport. Following hatching, larvae exhibit high aerobic capacity and elite swimming performance during the pelagic phase. As settlement approaches, however, maximum metabolic rate declines while hypoxia tolerance improves markedly. Transcriptomic analyses reveal coordinated shifts in haemoglobin subunit expression and increased expression of neuroglobin, cytoglobin, and myoglobin, consistent with developmental modulation of oxygen-binding properties. Concurrently, mitochondrial volume density peaks during metamorphosis, suggesting stage-specific reorganisation of oxidative capacity as larvae transition from sustained pelagic swimming to reef-associated life. Together, these results indicate that oxygen transport and cellular aerobic machinery are dynamically rebalanced across ontogeny to meet shifting ecological demands. By integrating cardio-respiratory development, globin expression, and mitochondrial restructuring, this work extends classical concepts of fish respiratory physiology into early life stages and tropical systems.

RESPIRATION Respiratory gas exchange and circulation: The legacy of Dave Randall

Importance of Blood (Tissue) Oxygen Extraction to the Environmental Tolerances of Fishes

Gamperl, Anthony (*kgamperl@mun.ca*) Oral Presentation
Co-Author: Porter, Emma and Durnford, Fiona
Memorial University of Newfoundland and Labrador

Abstract: The prevailing dogma is that cardiac output () is the primary determinant of temperature-dependent increases in fish oxygen consumption ($\dot{M}O_2$), and thatcapacity functions [e.g. cardiac stroke volume, haematocit and blood (tissue) oxygen extraction ($EO_2 = \dot{M}O_2$ /)] play little to no role. However, it is now clear that this is not true for many species (i.e., American eel, sablefish, Atlantic salmon schoolmaster snapper and Nile tilapia etc.), and that changes in EO_2 can be just as, or more, important in meeting the metabolic demands of fishes at high temperatures. Further, recent experiments reveal novel and important information about the interrelationships between , EO_2 and $\dot{M}O_2$ under various challenges. For example, while chronic hypoxia limits the pumping capacity of the trout heart, $\dot{M}O_2$ is not lower during a CTMax test, because EO_2 completely compensates. Warm temperatures, prior to severe hypoxia, reduce the salmon's capacity to increase EO_2 and hypoxia tolerance due to higher baseline (normoxic) values. When hypoxic salmon are warmed, increases in are minimal (and not correlated with changes in $\dot{M}O_2$), whereas changes in EO_2 are substantial and positively related to MO_2. Finally, in tilapia, EO_2 is critical to maintaining $\dot{M}O_2$ as they approach their lower thermal limit, and to continued increases in $\dot{M}O_2$ during a CTMax test after plateaus. This research shows that changes in EO_2 are a critical component of the fish's response to climate change-relevant stressors, and asserts that understanding interspecific differences in EO_2, and the mechanisms involved, should be a priority in the field of fish cardiorespiratory physiology.

RESPIRATION Respiratory gas exchange and circulation: The legacy of Dave Randall

Evolution of Respiratory Tolerance to Suspended Sediment in Freshwater Fishes

Koseck, Benjamin (*bkoseck@uoguelph.ca*) Oral Presentation
Co-Author: Turko, Andy and Young-Veenstra, Sarah
University of Guelph

Abstract: Suspended sediment is a stressor in aquatic habitats, which can abrade and clog fish gills. The respiratory impacts of sediment vary among fishes, but the mechanisms underlying these inter-specific differences are unknown. One idea, the lamellar spacing hypothesis, suggests that sediment-tolerant species have evolved greater inter-lamellar spacing that reduces clogging and maintains oxygen uptake. Another idea, the epithelial thickness hypothesis, posits that tolerant species have evolved a thicker lamellar epithelium to protect against abrasive damage. To test these hypotheses, we used a phylogenetic comparative approach and assessed sediment tolerance and gill morphometrics in 20 freshwater fishes. Sediment tolerance of each species was quantified by comparing maximum metabolic rate in fish acutely exposed to suspended sediment

(200 mg/L bentonite) versus a clean water control. Phylogenetically informed analyses indicated that sediment tolerance was negatively correlated with maximum metabolic rate, suggesting that species with greater aerobic capacities may face greater respiratory impairment from sediment. Furthermore, sediment tolerance was not phylogenetically conserved but rather appears to have evolved independently across lineages. Plasma [Cl-] is currently being analyzed to assess osmoregulatory impairment and gill epithelial integrity, and gill morphometrics such as inter-lamellar spacing and epithelial thickness are being quantified histologically to assess whether structural gill traits further explain interspecific differences in tolerance.By identifying mechanisms of sediment tolerance, this study provides insight into how fish have adapted to elevated suspended sediment levels and may also guide conservation programs by allowing us to predict species vulnerability.

RESPIRATION Respiratory gas exchange and circulation: The legacy of Dave Randall

Respiratory physiology in the era of climate change

Esbaugh, Andrew (*a.esbaugh@austin.utexas.edu*) Oral Presentation
University of Texas

Abstract: Ocean warming, acidification and deoxygenation are three of the most prominent environmental concerns facing global oceans. These stressors are linked not only by their connection to anthropogenically-driven climate change, but also by the fact that their modes of action involve respiratory gas exchange with subsequent implications for whole animal performance and metabolism. These connections serve as the basis for numerous frameworks that attempt to predict the long-term outcomes of climate change on fishes, while also acting as the building blocks for modern ecophysiological approaches. A particularly relevant area of research in the era of climate change is the exploration of adaptive capacity, which can refer to local adaptation and/or phenotypic plasticity. Many fish species endemic to habitats with naturally fluctuating abiotic conditions (e.g. estuaries) have the ability to dynamically control parts of their cardiorespiratory physiology to mitigate the effects of prolonged hypoxia, hypercapnia and warming – sometimes resulting in unintended physiological consequences. Crucially, the ability to develop strong, mechanistically-derived hypotheses in these areas requires a thorough understanding of foundational physiological concepts and their interactions with environmental factors. For fish physiologists, much of this foundational research – particularly in the fields of cardiorespiratory, acid-base and osmoregulatory physiology – was undertaken by the late Dave Randall, his students and collaborators. This presentation will explore the respiratory impairment and plasticity of estuarine fishes associated with climate change stressors, while drawing specific links to the work of Dave Randall in the fields of respiratory and environmental physiology.

RESPIRATION Respiratory gas exchange and circulation: The legacy of Dave Randall

Interactive effects of temperature and deoxygenation on the acclimation capacity of Atlantic croaker (Micropogonias undulatus).

Hancock, Grace (*gmhancock@vims.edu*) Oral Presentation
Co-Authors: Dichiera, Angelina
Virginia Institute of Marine Science

Abstract: Extreme environmental conditions are increasing in severity, frequency, and duration in coastal habitats. In Chesapeake Bay, a vital fish nursery habitat, heatwaves and hypoxic events are becoming common occurrences, which are likely to impact to the health of individuals and populations alike. While the physiological impacts of high temperatures and deoxygenation on fish are well understood when studied in isolation, fewer studies have evaluated the effects of simultaneous exposure. Those that have, found highly species-dependent and complex results when compared to a single stressor exposure. The aim of this project is to characterize the acclimation capacity of Atlantic croaker (Micropogonias undulatus) and determine the underlying mechanisms that may allow them to cope with warming and low oxygen over time. A sequential factorial design will be used to expose Atlantic croaker to a combination of environmentally relevant temperatures and oxygen levels (22 or 28°C, 100 or 70% dissolved oxygen) in which they will be tested repeatedly after acute and prolonged exposures (5 days and 4 weeks). Fish from each treatment will be tested for either their thermal tolerance (critical thermal maxima; CTmax) or their hypoxia tolerance (critical oxygen tension; Pcrit) and metabolic phenotype (aerobic scope, maximum metabolic rate, standard metabolic rate, and excessive post exercise oxygen consumption) using static intermittent-flow respirometry. Combined stressors are expected to have a synergistic effect on Atlantic croaker thermal and hypoxia tolerances, decreasing tolerance with acute exposure. However, with long-term exposure, fish are expected to improve tolerances, together with changes in oxygen extraction, transport, and demand.

RESPIRATION Respiratory gas exchange and circulation: The legacy of Dave Randall

Legacy effects of fishing on the climate resilience of fish

Goodrich, Harriet (*harriet.goodrich@utas.edu.au*) Oral Presentation
Co-Author: Rossiter-Hill, Finlay; Wolfe, Barrett; Audzijonyte, Asta; Breslin, Rachel and Tracey, Sean
Institute for Marine and Antarctic Studies

Abstract: In a warming and more variable ocean, fish populations persist partly because they contain a diversity of physiological types. Some individuals maintain performance across heat and low oxygen, others do not, and this variation buffers populations when conditions fluctuate. Fishing mortality erodes that diversity. Across regions and gear types, harvest consistently targets particular phenotypes, including larger, faster-growing, and bolder individuals, traits often associated with higher metabolic performance and elevated aerobic capacity. Over generations, this directional removal reshapes trait distributions

through fisheries-induced evolution, narrowing phenotypic variation and reducing the standing genetic and physiological diversity that underpins adaptive capacity. Although the interaction between fishing and climate change is widely discussed, few studies have directly tested whether exploitation alters physiological trait distributions or modifies responses to environmental extremes in wild populations, despite physiology placing fundamental constraints on thermal and hypoxia tolerance. To address this gap, I draw on a case study from Tasmania, Australia. The sand flathead (Platycephalus bassensis) is the state's most popular recreational finfish and occurs across a well-characterised gradient of harvest intensity. Across this gradient, we observe consistent regional divergence in growth, size-at-age, thermal tolerance, and postprandial metabolic performance. These patterns are consistent with long-term selective harvest and indicate that populations exposed to sustained fishing pressure may differ in their physiological responses to environmental extremes.

RESPIRATION Respiratory gas exchange and circulation: The legacy of Dave Randall

To boldly go… to the surface: linking use of aquatic surface respiration to personality and hypoxia tolerance in zebrafish (Danio rerio)

Flear, Kaitlyn (*kflea045@uottawa.ca*) Oral Presentation
Co-Author: Rabay, Joelle; Gilmour, Kathleen
University of Ottawa

Abstract: Although intraspecific variation in hypoxia tolerance is well documented, the factors underlying this variation have received less attention. Fishes mount physiological responses to hypoxia but may also respond behaviourally, for example, via aquatic surface respiration (ASR). Fish of low hypoxia tolerance rely more heavily on ASR during hypoxia – for example, zebrafish lacking Hif-1a initiated ASR at a higher water PO2 and spent more time performing ASR than wildtype fish. Performance of ASR also depends on perceived risk, suggesting that personality may impact its use at the individual level. To determine the relationships between personality, ASR use, and hypoxia tolerance in zebrafish, we measured the PO2 threshold of ASR initiation and ASR use during hypoxia, boldness (via the novel tank dive test, NTDT, and emergence test), and hypoxia tolerance, as time to loss of equilibrium (LOE). The repeatability of hypoxia tolerance, NTDT, and emergence tests was assessed. Because interspecific differences in behaviour during hypoxia are well known, individual behaviour was assessed during time-to-LOE trials. We predicted that fish with high time-to-LOE (high hypoxia tolerance) would make less use of ASR and be risk-averse. We found hypoxia tolerance to be moderately-to-highly repeatable in zebrafish, with time-to-LOE in females but not males dependant on food intake and season. Time to emergence was repeatable in both sexes, however behaviour in the NTDT was generally repeatable only in males. In contrast to our prediction, ASR use did not appear to be correlated with time-to-LOE.

RESPIRATION Respiratory gas exchange and circulation: The legacy of Dave Randall

When Do Plecos Breathe Air? Developmental Emergence and Environmental Drivers of Aerial Respiration in Ancistrus cirrhosis

Dzialowski, Edward (*ed.dzialowski@unt.edu*) Oral Presentation
Co-Author: Crowder, Lauren; Valdez, Hayley
University of North Texas

Abstract: The bristlenose pleco (Ancistrus cirrhosus), a loricariid armored catfish, breathes air using its highly vascularized stomach when exposed to hypoxic aquatic environments. This study examined the developmental onset of air breathing and the influence of temperature and aquatic oxygen availability on air-breathing behavior from juveniles to adults. To determine the timing of air-breathing onset, juveniles (1–8 cm total length) were exposed to a gradual decrease in aquatic oxygen from 100% to 8% air saturation over 50 minutes. Juveniles first breathed air between 2.1 and 3 cm, and all fish larger than 3 cm breathed air when challenged with hypoxia. To evaluate responses to temperature and oxygen availability, fish (2–14 cm) were exposed to three temperatures (20°C, 25°C, 30°C) and progressive hypoxia (100%, 30%, 15% air saturation). Gill ventilation frequency (Fg) and air-breathing frequency (Fabo) were monitored hourly at each temperature–oxygen combination. In normoxia, increasing temperature from 20°C to 30°C increased Fabo from 0.6 to 8.5 breaths hr^{-1}. At all temperatures, hypoxia significantly increased Fabo; at 15% air saturation fish increased Fabo to 6.9 breaths hr^{-1} at 20°C and 22.5 breaths hr^{-1} at 30°C. Fg increased to a lesser extent than Fabo with increasing temperature across oxygen levels. Developing A. cirrhosus begin aerial respiration near 3 cm, and increased oxygen demand at higher temperatures is met primarily through increased Fg rather than Fabo.

RESPIRATION Respiratory gas exchange and circulation: The legacy of Dave Randall

Zebrafish Biology: Ignore Sex at Your Peril

Burggren, Warren[1] (*burggren@unt.edu*) Oral Presentation
Co-Author: Martinez Bautista, Gil[1]; Hamilton, Trevor[2]
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Abstract: Zebrafish (Danio rerio) serve as a premier vertebrate model in biomedical, toxicological, and physiological research, yet sex differences are chronically underreported, compromising data reproducibility. This presentation, urging sex as a mandatory biological variable to report, synthesizes extensive evidence of sex-related differences across molecular, morphological, physiological, behavioral, and ecological domains in zebrafish and other fishes. PubMed analyses reveal <12% of zebrafish papers mention "male" or "female," far below rates for mammalian models (~30-50%). Comprehensive proteomics and transcriptomics uncover profound sex biases: e.g. female and male livers prioritize different anabolic and catabolic pathways. As another example, estrogen impairs male cardiac ejection fraction but enhances female cardiac output; aged females show distinctly different heart filling dynamics. Metabolic patterns reverse with age – as juveniles males zebrafish have higher metabolic rates than females, but this

pattern reverses in adults. Behaviorally, males display greater boldness and locomotion in youth, females later in life. Toxicology reveals sex-specific vulnerabilities, e.g., ethanol reduces locomotion in males, but shows paradoxical mixed stimulatory/depressant effects in females. Comparative data from tilapia, salmon, and medaka also highlight sexual differences in growth rate and pollution responses. Summarizing, ignoring sex in studies on zebrafish and other species inflates variability, misleads data interpretation, and hampers evolutionary insights, to mention a few negative implications. We recommend that investigators report strain/age, use two-way ANOVA (sex × treatment), and power studies for interactions. Embracing sex differences will enhance rigor, from basic physiology to translational applications.

POLAR The physiology, biology and ecology of polar fish

The physiology, biology and ecology of polar fish

Pedro Guerreiro and Luis Vargas-Chacoff

Abstract: Recent technological developments allowed to research deeper into the molecular and functional aspects of the biology and the phylogeny of these fish, and all this research is taking place at a time in which the future of these species is uncertain due to climate change. In the arctic there is novel information on the distribution of species due to temperature shifts, and in the Antarctic current studies focus very much on how the physiology adjusted to extreme environments and whether there is scope to acclimate to new conditions. Meanwhile emerging pollutants are starting to be found in these pristine regions, with likely impacts on fish. Icefish nesting grounds have recently been observed for the first time, new information about larval and early development are finally being looked at, and many questions about the antifreeze proteins are finding their answers.

POLAR The physiology, biology and ecology of polar fish

Temperature sensing channels in Arctic gadids

York, Julia (*jmy@illinois.edu*) Oral Presentation
Co-Authors: Cheng, C.-H. Christina
University of Illinois Urbana-Champaign

Abstract: Arctic fishes are experiencing accelerating ocean warming. Species with limited heat tolerance are expected to seek thermal refuge as a primary response. This requires that fishes sense temperature environments accurately. Further, any adaptive shift in thermal tolerance must include recalibration of the sensory system. Yet very little is known about the temperature sensing systems of fishes, their evolutionary capacity or constraints, and these questions have not been studied at all in Arctic fishes. In this study, we compared three Arctic gadids that vary in thermal tolerance: eurythermal Pacific cod (Gadus macrocephalus), intermediate saffron cod (Eleginus gracilis), and stenothermal polar cod (Boreogadus saida). Fishes were captured at summer temperatures (range 4-12°C) around coastal Alaska. High molecular weight DNA was extracted from blood samples, and long-read genomes were assembled for both species using a combination of PacBio HiFi reads (50x coverage), Hi-C scaffolding, and avidite sequencing short reads (40x coverage). Genome assemblies generated were ~700 Mbp in length and contained approximately 20% simple dinucleotide repeats, an extensive simple tandem repeat content that appears to be characteristic of Arctic gadids but imposes multiple challenges for genomic assembly algorithms. Putative thermosensory and non-sensory nervous and somatic tissues were dissected and sequenced using PacBio IsoSeq and Illumina reads. Expression of molecular mechanisms of temperature sensing, such as transient receptor potential (TRP) channels, will be compared. Proteins of interest will be structurally compared for evidence of adaptive change in the coding sequences. We hypothesize that thermosensory mechanisms will evolve first by regulatory, then structural evolution.

POLAR The physiology, biology and ecology of polar fish

Assessing fish community structure beneath the perennial ice-covered McMurdo Sound, Antarctica by environmental DNA metagenomics

Kim, Jinmu (*kjm04@illinois.edu*) Oral Presentation
Co-Authors: Cheng, C.-H. Christina
University of Illinois Urbana-Champaign

Abstract: McMurdo Sound, Antarctica, is a perennially ice-covered environment, precluding conventional underwater biodiversity surveys such as netting and trawling. We took the alternate approach of sampling seawater from multiple locations and depths across the Sound to recover environmental DNA (eDNA) and applied shotgun metagenomics sequencing and analyses to characterize the metazoan faunal diversity. Taxonomic classification analyses of the recovered metazoan eDNA identified nine phyla, 24 classes, 34 orders, 50 families, and 60 genera. Chordata was the predominant phylum, represented primarily by the class Actinopteri, enabling detection of rare or cryptic taxa often missed by traditional surveys. The genus Dissostichus was highly abundant within Perciformes, corroborating the seasonal (Austral summer) migration of large numbers of the giant Antarctic toothfish, Dissostichus mawsoni, into the Sound, which coincided with the time of water sampling. When Dissostichus sequences were excluded, the underlying ichthyofauna became apparent, with Trematomus, Notothenia, and Lycodichthys emerging as the most prominent genera. The beta-diversity analysis of Perciformes communities revealed clear depth-structured assemblages where shallow-water sites clustered together and were distinctly separated from deeper sites, supported by a significant depth-dissimilarity correlation. This study is the first attempt at unveiling the broader biodiversity under the ice-covered McMurdo Sound using eDNA methods. The findings demonstrate that eDNA metagenomics is an effective tool for assessing biodiversity in difficult-to-access polar environments and highlights the ability to resolve spatial patterns in fish community structure. However, limitations from incomplete metazoan reference databases or trace-level cross-ecosystem contaminations inevitably exist, which can affect low-abundance detections.

POLAR The physiology, biology and ecology of polar fish

Short-term resilience with hidden costs: physiological and molecular responses of juvenile Notothenia rossii to simulated marine heatwaves

Guerreiro, Pedro M.[1] (*pmgg@ualg.pt*) Oral Presentation
Co-Authors: Amaral, Inês[1]; Costa, Rita A.[1]; Gregorio, Silvia F.[1]; Nualart Carrasco, Daniela Paz[2]; Canário, Adelino V. M.[1]; Paschke La Manna, Kurt[2] and Vargas-Chacoff, Luis[2]
Centre of Marine Sciences CCMAR/CIMAR LA, [2]Universidad Austral de Chile

Abstract: Marine heatwaves are becoming more frequent and intense, posing a particular threat to cold-specialized Antarctic fishes. We investigated the physiological and

molecular responses of juvenile Notothenia rossii to simulated marine heatwaves (MHWs), with emphasis on both acute tolerance and post-stress recovery. Juveniles collected in the lower intertidal region were first assessed for critical thermal maximum (CTmax) using a dynamic ramping protocol, which established an upper tolerance range of 12.7–16.7 °C. In a second experiment, fish were gradually (3ºC/day) acclimated and exposed for five days to 5, 8, or 11 °C, or to the same warming protocol but followed by recovery at 2 °C. We measured oxygen consumption, plasma biomarkers, muscle glycogen, liver oxidative stress indicators, and tissue-specific expression of stress-related genes in brain, liver, gills, and heart. All fish survived up to 11 °C, indicating a degree of short-term tolerance to acute warming. Oxygen consumption increased markedly at 8 and 11 °C, but returned to baseline after recovery, showing reversible yet energetically costly metabolic adjustment. In contrast, oxidative damage was not fully resolved during recovery, with delayed lipid peroxidation and other temperature-dependent shifts in plasma and hepatic biomarkers. Muscle glycogen was largely preserved and rapidly restored after re-cooling. At the molecular level, classical heat shock responses were weak, whereas antioxidant and metabolic genes showed tissue-specific regulation, including cardiac patterns consistent with shifts in mitochondrial function and substrate use. Overall, juvenile N. rossii display moderate short-term plasticity, but recovery carries physiological costs that may reduce resilience to more frequent or prolonged marine heatwaves.

POLAR The physiology, biology and ecology of polar fish

Photoperiod–temperature mismatch has impacts on feeding, metabolism, and thermal tolerance in the Antarctic spiny plunderfish Harpagifer antarcticus

Guerreiro, Pedro M[1] (*pmgg@ualg.pt*) Oral Presentation
Co-Author: Vargas-Chacoff, Luis[2]; Nualart Carrasco, Daniela Paz[2]; Costa, Rita A.[1]; Paschke La Manna, Kurt[2]; Rosas, Carlos[3]; Canário, Adelino V. M.[1]; McCormick, Stephen D.[4] and Brazão, João[1]
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Abstract: Organisms rely strongly on seasonal environmental cues, particularly photoperiod and temperature, to synchronize physiological processes with predictable annual cycles. In polar regions, photoperiod provides a highly reliable signal, whereas temperature exhibits relatively small but seasonally structured variation. Climate change is altering the magnitude and timing of thermal conditions, potentially uncoupling these cues and exposing organisms to novel combinations of daylength and temperature. We investigated how photoperiod–temperature asynchrony affects physiological performance in the Antarctic spiny plunderfish Harpagifer antarcticus. Fish were exposed to simulated summer photoperiod (20 h light:4 h dark) or winter photoperiod (4 h light:20 h dark) combined with present-day summer temperature (2–3 °C) or elevated temperature (5–6 °C). Fish were sampled after 9, 18, and 36 days of exposure. Physiological performance was evaluated through measurements of feeding intake (amphipod consumption), condition factor, metabolic rates during light and dark periods, thermal sensitivity (Q10),

and critical thermal maximum (CTmax). Activities of oxidative stress enzymes in liver tissue were also quantified to assess potential cellular responses. Photoperiod influenced several aspects of physiological performance. Feeding intake and condition factor differed between photoperiod treatments, indicating that seasonal light regimes influence energy acquisition and overall physiological status. Temperature was the primary driver of metabolic rate; however, photoperiod modulated these responses, with higher thermal sensitivity (Q10) observed under short photoperiods, particularly during dark periods. Critical thermal maximum showed modest variation among treatments, suggesting that seasonal light conditions can influence thermal tolerance even in highly cold-adapted Antarctic fishes. Enzymatic indicators of oxidative stress also varied among treatments, indicating potential adjustments in cellular defence mechanisms. These results suggest that photoperiod contributes to seasonal physiological programming and can modify how Antarctic fishes respond to warming. Climate-driven disruption of the natural coupling between temperature and photoperiod may therefore influence the physiological performance and resilience of polar fishes under future environmental change.

POLAR The physiology, biology and ecology of polar fish

Impact of microplastic PVC and thermal stress on fish microbiomes and cellular stress genes in Antarctic and sub-Antarctic fish.

Nualart Carrasco, Daniela Paz[1] (*daniela.nualart@gmail.com*) Oral Presentation
Co-Authors: Schwob, Guillaume[2]; Orlando, Julieta[3]; Paschke La Manna, Kurt[1]; Guerreiro, Pedro[4]; McCormick, Stephen D.[5]; Cheng, C.-H. Christina[6] and Vargas-Chacoff, Luis[1]
[1]Universidad Austral de Chile, [2]Université Claude Bernard Lyon 1; [3]Universidad de Chile, [4]Centro de Ciências do Mar, [5]University of Massachusetts, [6]University of Illinois Urbana-Champaign

Abstract: Antarctic and sub-Antarctic fishes of the genus Harpagifer inhabit extreme environments and face emerging anthropogenic stressors, including plastic pollution and warming. While each factor is known to affect fish physiology, their combined impacts on molecular responses and host–microbe interactions remain poorly understood. This study investigates the microbial and physiological responses of two notothenioid species, the Antarctic Harpagifer antarcticus and the Sub-Antarctic Harpagifer bispinis, to the combined challenge of PVC microplastics and thermal increase. Fish were exposed to PVC microplastics through acute oral administration, under thermal regimes: current ambient temperatures (2°C/8°C) and projected 2050 climate change scenarios (5°C/12°C) for 24 h respectively. We evaluated gut microbiome composition using 16S rRNA sequencing and gene expression of cellular stress genes (RT-qPCR). Our findings reveal that the diversity of gut microbiomes in both species declined significantly under the combined stress of PVC and elevated temperatures. This microbiome shift was accompanied by a marked alteration in the expression of cellular stress genes, including heat shock protein 70 (hsp70), S100 calcium-binding protein (s100), High Mobility Group 1 box (hmg1b), E3 Ubiquitin Ligases (E3), and BCL2-associated X (Bax). These results demonstrate that, while notothenioids exhibit plastic responses to individual stressors, the combined effects of

warming waters and plastic pollution lead to microbiome dysbiosis and compromised cellular stress genes. This research provides essential baseline data regarding the resilience of Southern Ocean ichthyofauna in an increasingly anthropized environment. This work was supported by Fondecyt 1250678 (LVCh), ANID-Millennium Science Initiative Program-Center ICN2021_002 and INACH.

POLAR The physiology, biology and ecology of polar fish

Antarctic fish as a novel source for age-related disease therapeutics: KOPRI's bioprospecting initiative

Kim, Jin-Hyoung[1] (*kimjh@kopri.re.kr*) Oral Presentation

Co-Authors: Kim, IL-Chan[1] and Yoon, Mee-Sup[2]

[1]Korea Polar Bear Research Institute, [2]Gachon University

Abstract: Notothenioid fishes of Antarctica present a biological paradox relevant to aging research: under environmental conditions that would accelerate cellular senescence in most vertebrates — chronic hypoxic stress, near-freezing temperatures, and extreme metabolic constraint — these species maintain constitutively active mTOR anabolic signaling, maximized mitochondrial biogenesis, and exceptional antioxidant homeostasis. Their genome and metabolome effectively encode a sustained resistance to the molecular hallmarks of aging, making them an evolutionary model of cellular resilience rather than merely an ecological curiosity. The Korea Polar Research Institute (KOPRI) is translating this biological archive into a first-in-class drug discovery program for age-related diseases. The initiative targets the conserved upstream drivers shared across sarcopenia, neurodegeneration, and metabolic decline — mitochondrial dysfunction and proteostatic failure — rather than pursuing disease-specific downstream symptoms. Sarcopenia serves as the clinical entry point: a condition with formal disease classification yet zero approved pharmacotherapies, for which a lead compound has already been secured. The same mechanistic pathways extend directly into Alzheimer's and Parkinson's disease pipelines. KOPRI's distinctive competitive advantage lies in custodianship of the world's sole comprehensive polar organism genomic and metabolomic database, now integrated with AI-driven compound discovery and predictive ADME modeling. This positions KOPRI at a singular intersection: between nature's most stringent evolutionary laboratory and the urgent pharmacological demands of aging global populations.

POLAR The physiology, biology and ecology of polar fish

Compound effects of warming, hypoxia, and salinity on metabolic physiology in the Antarctic coastal fish Harpagifer antarcticus

Guerreiro, Pedro M.[1] (*pmgg@ualg.pt*) Poster Presentation
Co-Authors: Costa, Rita A.[1]; Nualart Carrasco, Daniela Paz[2]; Teixeira, José P. P.[1]; Paredes, Danae[2]; Vargas-Chacoff, Luis[2] and Paschke La Manna, Kurt[2]
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Abstract: The Antarctic spiny plunderfish Harpagifer antarcticus is a small benthic notothenioid inhabiting shallow coastal habitats along the Antarctic Peninsula and surrounding archipelagos. Its sedentary behaviour, strong site fidelity, and occurrence in nearshore environments suggest that individuals may experience substantial local environmental variability. However, little is known about the metabolic limits of this species or how multiple environmental drivers interact to influence physiological performance. In particular, warming, reduced oxygen availability, and salinity changes may jointly affect respiratory and osmoregulatory processes in coastal Antarctic fishes.
We examined metabolic responses of H. antarcticus using static and intermittent respirometry across a series of environmental manipulations. Fish (3–30 g) were acclimated to 3, 6, and 10 °C, and oxygen consumption was measured to estimate routine metabolic rates and thermal sensitivity (Q10). Critical oxygen thresholds were determined by progressively reducing oxygen saturation from normoxia to severe hypoxia. To explore interactions among environmental drivers, fish were also acclimated to combinations of temperature (2 or 6 °C) and salinity (35 or 12.5 psu) before measurements of metabolic rates under normoxic and hypoxic conditions.
H. antarcticus tolerated temperatures exceeding its typical environmental range (2–4 °C), but warming resulted in increased oxygen demand. Routine metabolic rates increased markedly with temperature, with Q10 values indicating strong thermal sensitivity. Metabolic scaling relationships varied with body size, with larger individuals showing steeper increases in oxygen consumption at elevated temperatures. Hypoxia trials revealed reductions in metabolic rates below critical oxygen levels with limited size dependence. Combined temperature–salinity–oxygen experiments suggest that osmotic conditions may influence metabolic responses to oxygen limitation, warranting investigation into osmorespiratory compromise.
These findings highlight the metabolic sensitivity of H. antarcticus to warming and oxygen availability and emphasize the importance of considering interacting environmental drivers when assessing the vulnerability of Antarctic coastal fishes to environmental change.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Environmental physiology of life-history transitions: A tribute to Steve McCormick

Ciaran Shaughnessy, Jill Leonard and Jason Breves

Abstract: This symposium will celebrate the scientific legacy, community impact, and recent retirement of Dr. Stephen D. McCormick. His pioneering work in fish physiology (especially in osmoregulation, endocrinology, and smolt physiology) and his stewardship of the community of fish physiologists (including his role as President of AFS Physiology) have shaped our field for over four decades. In brief, Steve's work has provided new insights into how environmental factors such as endocrine disrupters, acidification, and climate change impact the performance (survival, growth, and reproductive success) of wild fish stocks including Atlantic salmon, American shad, blueback herring, brook trout, alewife, Atlantic sturgeon, and sea lamprey. Moreover, Steve's applied work with hatcheries and dams relied on the discoveries he made regarding the hormonal control of osmoregulatory systems and the effects of environmental temperature fluctuations on hormonal and cellular stress responses. This symposium will bring together Steve's trainees, collaborators, and colleagues from around the world to share their latest research findings and reflect on how their career and research are intertwined with Steve's scholarship, mentorship, and friendship. The talks will cover a range of taxa, analytical methods, and physiological systems, reflecting the integrative and interdisciplinary spirit that defined Steve's career. This symposium showcases the ongoing ripple effects of his science and community-building, celebrating not only Steve's scientific career, but also the vibrant scientific "school" of fish physiologists he helped cultivate

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IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

The light-pituitary TSH/brain DIO2/T3 pathway for stimulating plasma GH and smoltification in juvenile Atlantic salmon

Björnsson, Björn Thrandur[1] (*thrandur.bjornsson@bioenv.gu.se*) Oral Presentation
Co-Author: McCormick, Stephen D.[2]
[1]University of Gothenburg; [2]University of Massachusetts Amherst

Abstract: Over the last 40 years, the collaborating labs of McCormick and Björnsson have studied the physiology and endocrine regulation of Atlantic salmon smoltification, together with numerous collaborators. Among their key findings is that increased daylength triggers elevated plasma GH levels in FW-living parr, leading to physiological changes that are adaptive for marine survival and establishing GH as a main driver of salmon smoltification. The light-induced regulatory pathways of GH elevation have, however, not been well elucidated. Our recent studies establish that increase in daylength results in increased pituitary tshβb mRNA levels in salmon parr, together with increased plasma GH, gill NKA activity and gill. In vivo exposure to long days causes increased dio2b transcription and T3

levels throughout the brain. Further, isolated (in vitro) pituitaries from juvenile salmon directly respond to increased daylength with elevated tshβb transcription. As intracerebral T3 treatment elevates plasma GH levels, the data suggest a pathway where photoperiod acts directly at the level of the pituitary, elevating TSH secretion, central DIO2 and T3 levels, leading to elevated plasma GH levels which drives the parr-smolt transformation. Studies are underway examining if TSH may also have direct stimulatory effect on GH secretion in the salmon pituitary.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Regulatory Plasticity of New Salmonid Na^+/K^+-ATPase α-subunit Paralogs and Functional Compensation in nkaα1b1/b2 Crispants

Nilsen, Tom Ole (*Tom.Nilsen@uib.no*) Oral Presentation
University of Bergen

Abstract: Na+, K+-ATPase (NKA) enzyme activity increases in osmoregulatory organs is associated with switching between the freshwater “Nkaα1a” and seawater “Nkaα1b” during parr-smolt transformation (PST) and salinity acclimation. They are also primarily located in “Nkaα1a” and “Nkaα1b” type ionocytes. Here we show that the “α1b” and “α1a” are encoded by paralogue genes that are not the result of Ss4R but of a duplication in a common ancestor of salmonids and esociformes and thus renamed nkaα1aa and nkaα1ab as they both are orthologues of nkaα1a in other teleosts. The Ss4R paralogues (nkaα1aa1, -1aa2, -1ab1, and -1ab2) was identified and found to be differentially regulated in response to photoperiod and increased salinity. The orthologue of teleost nkaα1b, called nkaα1c in salmonids also possesses differently regulated Ss4R paralogues (nkaα1c1, -1c2). These six nkaα1 genes divide in three groups with a distinct trajectory that during PST. Crispr/Cas9 knock down of the "α1b" paralogs (nkaα1b1 and nkaα1b2) in Atlantic salmon resulted in compensatory increase in gene and protein abundance of “α1a” paralogs in nkaα1b1/b2 crispants. Immunohistochemistry revealed a increased presence of NKAα1a positive ionocytes in the gills of nkaα1b1/b2 crispants compared to controls, suggesting that nkaα1b1/b2-/- ionocytes in the mosaic nkaα1b1/b2 crispants may send out signals to compensate for the loss of nkaα1b1/b2. The mechanism behind this compensation remains unknown.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Comparative transcriptomics of three Atlantic salmon (Salmo salar) stocks sampled at West Greenland

Ellingson, Brandon[1] (*Brandon.ellingson@noaa.gov*) Oral Presentation
Co-Authors: Sheehan, Timothy[1]; Bradbury, Ian[2]; Speciale, Emily[3] and Shartau, Ryan[4]
[1]NOAA’s National Marine Fisheries Service, [2]Department of Fisheries and Oceans Canada, [3]The University of North Carolina at Chapel Hill, [4]Washington State University

Abstract: Contemporary Atlantic salmon populations in North America extend from Maine, USA to northern Québec, Canada. Despite large geographic separation, these populations migrate to common marine feeding grounds in the Labrador Sea and off the coast of Greenland. Ecosystem conditions are shared at marine feeding grounds, but individual stocks are subject to population-specific factors that may influence survival enroute. Marine mortality is the primary driver of range-wide declines in Atlantic salmon and it is important to better understand the interaction between population-specific factors influencing marine survival versus large scale common drivers. One approach is to evaluate stock-specific differential gene expression at shared feeding grounds which may suggest potential drivers of survival. Individual fish harvested off the coast of West Greenland were sampled for biological characteristics, assigned to a region of origin via genetic analysis and analyzed for differential gene expression using RNA-sequenced liver tissue. Results from 12 individuals equally representing three region of origin groups across the latitudinal North American range of the species (southern; United States, mid; Gaspe Peninsula and northern; Labrador) indicate that the majority of differentially expressed genes were related to protein synthesis, metabolism, the unfolded protein response, immune function and xenobiotic detoxification. The transcriptome profiles of the two southern groupings were indicative of lipid and protein catabolism, reduced growth, pollution exposure, elevated stress, and an altered immune response. The stock-specific gene expression characterized in this study may help to inform differences in marine survival that have been reported across these Atlantic salmon stocks.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

The Intestinal "Saga" started with Steve! or What is happening in the intestine during the parr-smolt transformation.

Sundell, Kristina Snuttan (*kristina.sundell@bioenv.gu.se*) Oral Presentation
Co-Author: Panagiotidis, Johannes; Hedfalk, Kristina; de Fonseka, Rane and Sundh, Henrik
University of Gothenburg

Abstract: The Atlantic salmon parr-smolt transformation is the most extensively studied pre-adaptive life history transition in fish. The importance of the physiological reorganization of the gills during parr-smolt transformation is undeniable, thanks to the work by Steve. During your visit to Gothenburg in late 80's you taught me the ATPase assays, and together we worked with the specific intestinal challenges. This marked the beginning of our continued work to understand the physiology of the intestinal hypo-osmoregulatory ability. During both natural and photoperiod-controlled smoltification, intestinal NKA activity increases, forming a broad peak that is elevated following seawater transfer. Immunohistochemistry confirms that enterocyte NKAs, the NKA-α1c isoform, increases in fresh water, initially displaying a basal localization that shifts to more basolateral distribution after seawater transfer. In parallel, NKCC(I) expression increases primarily with an apical localization. This coincides with enhanced active transport in the intestinal epithelia and a peak in the intestinal fluid transport capacity. During this period, epithelial permeability is also modified. Paracellular permeability decreases during the

smoltification which is followed by a rapid tightening of the paracellular pathway after seawater transfer. This suggests a redirection of fluid movement from a paracellular route in fresh water, to a transcellular route in seawater. Such a shift may be mediated by differential aquaporin expression, physiological regulation like trafficking or gating of aquaporins in the enterocyte membrane, or through the regulation of the fatty acid composition of the membrane phospholipids, which could alter the enterocyte membrane permeability to water.
Thank you, Steve, for supporting my “intestinal obsession”.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Prolactin promotes putative pathways for Cl- absorption in Atlantic salmon ionocytes

Breves, Jason[1] (*jbreves@skidmore.edu*) Oral Presentation
Co-Authors: Tao, Yixuan[1]; Regish, Amy[2] and McCormick, Stephen D.[3]
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Abstract: Although prolactin is known to facilitate the survival of teleosts in freshwater (FW) environments, the branchial mediators of this activity remain unresolved in salmonids. In this regard, progress has been hindered by a paucity of identified solute transporters that support Na+ and Cl- absorption in FW-type ionocytes. We used anadromous Atlantic salmon (Salmo salar) smolts to first examine how environmental salinity modulates the expression of genes encoding candidate pathways for branchial ion absorption, namely Na+/H+ exchangers (Nhes) and Clc-family Cl- channels (Clcs). We then tested the hypothesis that prolactin transcriptionally activates solute transporters that are upregulated under FW/ion-poor conditions.Among an array of Nhe- and Clc-encoding genes identified through phylogenetic analyses, nhe3.2, clc2a.1, clc2a.2, clc2b.2,and clc2b.3 showed higher expression in the gills of FW- versus seawater-acclimated smolts. Compared with smolts held under ‘standard’ FW conditions, smolts exposed to ‘ion-poor’ conditions showed enhanced expression of nhe3.2, clc2a.2, clc2b.2, and clc2b.3 in parallel with Na+/HCO3-cotransporter 1.2a, Na+/K+-ATPase-alpha1a, Cl-/HCO3-exchanger (slc26a6a2), and prolactin receptor a. The addition of ovine prolactin to incubated gill filaments stimulated the expression of clc2b.3 and slc26a6a2, but not of any other investigated gene transcripts.Our results indicate that prolactin signaling supports acclimation to FW/ion-poor environments by promoting a candidate Cl--uptake transport pathway that leverages Slc26a6a2 and Clc2b.3. Further study is now warranted to determine whether Slc26a6a2 and Clc2b.3 are co-expressed within the FW-type ionocytes of salmon.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Endocrine coordination of appetite and reproductive migration by dual ghrelin receptors in the Japanese eel

Tsukada, Takehiro[1] (*takehirotsukada@sci.toho-u.ac.jp*) Oral Presentation
Co-Authors: Yoshizawa, Aya[1]; Kano, Chiaki[1]; Kashima, Makoto[1]; Wong, Marty Kwok-Shing[2]; Yada, Takashi[3] and Kaiya, Hiroyuki[4]
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Abstract: The Japanese eel (Anguilla japonica) is a migratory fish that undertakes one of the longest oceanic spawning migrations among teleosts, comparable to salmon. During its freshwater growth phase, the eel is a top predator with a strong appetite necessary for increasing body mass to support ovarian and testicular development. In contrast, during spawning migration, eels abruptly cease feeding, accompanied by degeneration of the digestive tract and teeth, and swim over 2,500 km from the East Coast to the Mariana Trench. However, the involvement of feeding regulation in sexual maturation has not been fully elucidated. In this study, we focused on receptors for the appetite-inducing hormone ghrelin, known as growth hormone secretagogue receptors (GHSRs). We recently identified two GHSRs in the Japanese eel, designated as RDS-type and KET-type GHSRs according to their peptide motif signatures. Multiplex mRNA detection using in situ hybridization chain reaction revealed that the RDS-type GHSR is localized in agrp1-positive neurons in the hypothalamic arcuate nucleus, whereas the KET-type GHSR is specifically expressed in growth hormone–producing cells in the pituitary, suggesting that ghrelin regulates both appetite and growth hormone secretion. UMAP visualization of anatomical, endocrine, and gene expression data from 75 wild-caught female eels classified individuals into four maturation stages, with RDS-type expression associating with the growth phase while KET-type expression associating with the final maturation phase. These findings suggest that expression of the two GHSRs is functionally segregated, potentially switching the physiological priorities from body mass accumulation to preparation for spawning migration in the Japanese eel.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Stress and growth – is myostatin a link between these two?

Biga, Peggy[1] (*peggy.biga@txstate.edu*) Oral Presentation
Co-Author: McCormick, Stephen[2]; Galt, Nicholas[3]
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Abstract: Stress is one of the major factors negatively contributing to production and animal welfare in aquaculture systems. Stressors like handling, stocking density, transportation, and disease are inevitable, and if not managed properly can negatively impact growth, reproduction, immune function, and survival. The effects of stress on fish growth have been well studied, and elevated levels of the primary stress hormone in fish,

cortisol, are linked to protein degradation and the inhibition of skeletal muscle growth. These effects of cortisol likely elicit these effects in concert with suppression of growth hormone secretion and responsiveness, while increases in somatostatin and myostatin, both of which are negative regulators of growth. However, a link between cortisol and myostatin expression and function is not clear. We therefore sought to identify if crowding stress from increased stocking density in multiple salmonids affected skeletal muscle myostatin expression. We also evaluated the effects on HSP90, a key molecular chaperone crucial for cortisol mediated cellular action. This study demonstrated, for the first time, a link between an acute stress response and myostatin in multiple salmonids. In addition, the study provided valuable evidence supporting a divergence in myostatin regulation among salmonids, where a second genome duplication event resulted in multiple myostatin genes. There is still a need to directly test the direct interaction between cortisol and myostatin in these species to better understand specific-specific responses.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Metabolic reprogramming in zebrafish: an obesity model platform for bioactive extract bioprospecting.

Ferreira-Martins, Diogo (*dmartins@ciimar.up.pt*) Oral Presentation
Co-Authors: Queiroga, Inês; Silva, Maria; Moxo, Javier; Papadopoulos, Konstantinos; Peris, Joel and Urbatzka, Ralph
University of Porto

Abstract: The zebrafish (Danio rerio) is a vertebrate model for studying metabolic regulation due to conserved endocrine pathways, genetic accessibility, and suitability for in vivo screening. It provides translational insight into human metabolic disorders, including obesity. Microalgae, rich in bioactive metabolites, may modulate energy balance, lipid metabolism, and feeding regulation. Combining zebrafish assays with microalgal bioprospecting and obesity models enables identification of conserved metabolic modulators relevant to human health. Here, we demonstrate the potential of organic extracts derived from microalgae, as a source of novel bioactive compounds for advancing metabolic disease research. Using a transgenic zebrafish model [Tg(ins:GFP-T2A-NTR3, cryaa:mcherry)] we demonstrated that dichloromethane/methanol extracts from microalgae recovered pancreatic β-cells numbers after ablation. In wildtype zebrafish, aqueous extracts of microalgae lowered lipid accumulation and suppressed appetite. Ethanolic extracts increased glucose absorption and reduced/increased food intake. Bioactivity-guided molecular networking with liquid chromatography-tandem mass spectrometry and GNPS identified compounds responsible for bioactive properties. Adult zebrafish were used to develop diet-induced obesity models via high-fat and overfeeding regimens. Our data indicated overfeeding produced the most robust phenotype, corroborated fat quantification by micro-computed tomography. This optimized model was subsequently applied for in vivo evaluation of previously identified bioactive extracts. Our findings highlight microalgae as sources of metabolic modulators and establish zebrafish diet-induced obesity models as a platform for screening and in vivo validation of bioactive

compounds relevant to obesity research. This work was funded by National Funds through FCT - Fundação para a Ciência e a Tecnologia, I.P., under the scope of the project COMPETE2030-FEDER-00871800.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Thermal Tolerance in Chinook Salmon Varies with Life-History and Smoltification

Metzger, David [1] (*dmetzger@zoology.ubc.ca*) Oral Presentation
Co-Author: Healy, Timothy[2]; Wellband, Kyle[2]; Schulte, Patricia[1]
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Abstract: Rising river and ocean temperatures are reshaping the environments experienced by anadromous fishes. Smoltification, a hormonally mediated life-history transition that reorganizes osmoregulatory, endocrine, and cellular physiology in preparation for seawater entry, occurs during this period of environmental change and may influence how fish tolerate thermal stress. Yet how this physiological remodeling interacts with thermal tolerance is not well understood. We evaluated thermal tolerance in four populations of Chinook salmon (Oncorhynchus tshawytscha) representing stream-type and ocean-type life histories that differ in freshwater residency and timing of seawater entry. We measured critical thermal maxima (CTmax) in both freshwater and seawater and integrated these data with population-level genomic and transcriptomic analyses. Stream-type populations showed consistently less tolerant of an acute thermal ramp compared to ocean-type populations. Smoltification reduced thermal tolerance in all populations, revealing a physiological trade-off between seawater preparedness and acute thermal tolerance. Thermal tolerance was more variable in saltwater, underscoring the context dependence of phenotypic expression. Genomic analyses identified population-specific variants and distinct expression profiles associated with thermal performance. However, resilient individuals across all populations shared enrichment of common functional pathways, including the unfolded protein response and ion transport mechanisms. These findings suggest that while populations may rely on different regulatory architectures, they converge on similar core physiological processes to cope with thermal stress. Our results highlight the polygenic and context-dependent nature of thermal tolerance in Chinook salmon and demonstrate how life-history strategy, developmental transitions, and genomic background interact to shape thermal tolerance.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Acclimation to high salinity confers cross-tolerance to acute heat stress in Fundulus heteroclitus

Monette, Michelle[1] (*monettem@wcsu.edu*) Oral Presentation
Co-Author: Breves, Jason[2]
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Abstract: Cross-tolerance occurs when exposure to one stressor enhances tolerance to a second, unrelated stressor, often through activation of shared defense mechanisms or response pathways. We examined whether acclimation to high salinity confers cross-tolerance to heat stress in the common mummichog (Fundulus heteroclitus). Fish were acclimated to low (3 ppt) and high (60 ppt) salinity for at least four weeks, after which we measured upper thermal tolerance, indices of whole-animal homeostasis, and the expression of heat shock protein genes (hsp70-1, hsp70-2, and hsc70) before and after a 120-min heat shock. Critical thermal maximum was elevated (+1.3°C) in high-salinity fish compared with low-salinity fish, indicating improved upper thermal tolerance. However, high-salinity fish exhibited greater homeostatic disturbance following the 120-min heat shock, reflected by higher plasma osmolality and glucose levels. Elevated thermal tolerance in high-salinity fish was associated with higher baseline gene expression of a constitutive heat shock protein (hsc70) and the blunted upregulation of inducible heat shock proteins (hsp70-1 and hsp70-2) in both gill and heart tissues. Together, these findings suggest that acclimation to high salinity confers cross-tolerance to heat stress in mummichogs, as evidenced by elevated thermal limits and reduced cellular stress responses. However, cross-tolerance was associated with greater disruption of whole-animal homeostasis, suggesting a trade-off that may constrain other physiological systems. Future work should determine whether high salinities enhance only acute heat-stress tolerance or also improve long-term resilience, including recovery capacity and performance during repeated heat events.

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Photoperiod is controlling the Thyroid axis in the Antarctic fish Harpagifer antarcticus

Vargas-Chacoff, Luis[1] (*luis.vargas@uach.cl*) Oral Presentation
Co-Author: Nualart Carrasco, Daniela Paz[1]; Guerreiro, Pedro M.[2]; Paschke La Manna, Kurt[1]; McCormick, Stephen D.[3]
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Abstract: The Antarctic region has two very distinct seasons: the summer, peaking at nearly 24 hours of daylight, and a winter season with long periods of darkness. As in other latitudes, fish in Antarctica likely have a seasonal clock that signals the endocrine system, but this interaction has not been widely examined. Therefore, we set out to investigate how photoperiod regulates the thyroid axis in Harpagifer antarcticus, a model species for Antarctic fish. To do this, we exposed fish to photoperiods of either 20:4 or 4:20 (hours of

light:dark) at two temperatures. Additionally, in a parallel experiment, fish in those conditions were injected with triiodothyronine (T3), or with iopanoic acid (which inhibits the peripheral conversion of T4 to T3, reducing T3 availability). Results show that osmoregulation and metabolism were affected by photoperiod and manipulation of the thyroid axis. We also observed changes in the oxidative response, indicating that daylength or photoperiod may act to signal the cellular stress response.Photoperiod was a physiological regulator, allowing the fish to adapt their response to temperature change, the latter having clear impacts on metabolism and, therefore, on the seasonal physiology of Antarctic fish.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Interrelations of the growth hormone/insulin-like growth factor system and somatostatin associated with seawater adaptation of euryhaline fish

Sheridan, Mark (*mark.sheridan@ttu.edu*) Oral Presentation
Texas Tech University

Abstract: Growth hormone (GH) and insulin-like growth factor-1 (IGF-1) have been shown to increase seawater (SW) adaptability of euryhaline fish by altering the expression and activity of ion transporters. For example, mRNA expression of seawater (SW)-adapting ion transporters (e.g., NKAα1B, NKCC, and CFTR) increase during smoltification and SW exposure of juvenile Atlantic salmon in association with the activation of cell signaling pathways that mediate growth hormone (GH) action. In addition, SW exposure upregulates the GH/IGF system of trout and salmon, including increasing mRNA expression of GH receptors, IGF1, and IGF receptors. By contrast, somatostatin reduces SW adaptability by inhibiting elements of the GH/IGF system (e.g., plasma GH as well as GHR, IGF-1, and IGFR mRNA expression) and by blocking SW-associated upregulation of GH/IGF system elements. The directs effects of GH on SW adaptation and ion transporter expression in isolated rainbow trout gill filaments revealed the mechanisms of GH action. Notably, both SW and GH exposure increase the expression of NKAα1B (SW form), NKCC, and CFTR mRNAs, but decrease mRNA expression of NKA α1A (freshwater form). In addition, SW and GH expression activate the JAK, STAT, and Akt cell signaling elements, but there are no effects on ERK or PKC activation. Moreover, inhibition of the JAK-STAT and PI3K-Akt pathways block SW- and GH-induced expression of NKAα1b (SW form), NKCC, and CFTR. Taken together, the GH/IGF system and somatostatin antagonistically regulate SW adaptation in euryhaline fish by modulating osmoregulatory apparatus (NKA, NKCC, CFTR) through differential activation of the JAK-STAT and PI3K-Akt signaling pathways.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Prolactin signaling in the highly osmotolerant Mozambique tilapia, Oreochromis mossambicus

Seale, Andre (*seale@hawaii.edu*) Oral Presentation
Co-Authors: Cao, Ke[1]; Singh, Simran[1]; Scalambrino, Brooke[1]; Chang, Ryan[1]; Goodearly, Tyler[1]; Merlo, Reilly[1] and Breves, Jason[2]
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Abstract: Teleosts achieve hydromineral homeostasis by hormonally modulating ion transport mechanisms. In the euryhaline Mozambique tilapia (Oreochromis mossambicus),two prolactin (Prl) isoforms, Prl188 and Prl177, are released from the pituitary in response to hyposmotic stimulation to promote branchial ion absorption. These isoforms signal through two Prl receptors (Prlrs), Prlr1 and Prlr2, which are expressed in the pituitary and key ionoregulatory organs. To understand how Prl signaling operates across a range of salinities that reflect their scope for osmotolerance, we exposed Mozambique tilapia to conditions spanning from fresh water (FW; < 0.1‰) to triple-strength seawater (3x SW; 105‰). Moreover, we employed ratios of prls and prlrsto facilitate the interpretation of regulatory patterns in response to salinity challenges. In the pituitary, prl188/prl177 and prlr1/prlr2 ratios decreased as salinity increased, dropping to levels in 3x SW that were less than 10% of those in FW. Branchial prlr1/prlr2 ratios also decreased with increased salinity, in parallel with effectors of branchial ion uptake, including Na+/Cl- cotransporter 2and Na+/K+ ATPase-a1a. These shifting ratios reflect a coordinated downregulation of prlr1 and upregulation of prlr2 as salinity increases. Conversely, transcripts encoding mediators of ion extrusion, such as Na+/K+/2Cl- cotransporter 1a andcystic fibrosis transmembrane conductance regulator 1, were elevated by high-salinity conditions. Our findings indicate that when Mozambique tilapia are exposed to hypersaline conditions, Prl signaling shifts toward Prl177 and Prlr2 relative to Prl188 and Prlr1. This isoform-specific plasticity likely underpins the exceptional euryhalinity and osmotolerance of this species.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Life-history divergence in renal glomerular morphology of Alewife (Alosa pseudoharengus)

Schultz, Eric T.[1] (*eric.schultz@uconn.edu*) Oral Presentation
Co-Authors: Mouchlianitis, Foivos[2]; Thai, Brandon[3]; McCormick, Stephen D.[4]
[1]University of Connecticut; [2]Aristotle University of Thessaloniki, [3]University of Saint Joseph, [4]University of Massachusetts

Abstract: The requirement for water elimination in fishes is primarily managed by the kidneys and is greater in freshwater environments than in seawater. Hence microevolutionary changes in renal structure are expected when populations adapt to changes in salinity regime. Alewife provides an excellent system to examine such evolutionary responses as multiple landlocked populations have independently evolved from anadromous ancestors. We quantified renal glomerular morphology using

microphotographs of histological kidney sections from freshwater juveniles representing one anadromous and multiple landlocked populations from Connecticut, USA, predicting that the landlocked forms will possess more extensive area for filtration. Landlocked populations exhibited larger glomeruli compared to the anadromous one after accounting for body size, while glomerular density did not differ between the life history forms. These findings are consistent with previous investigations indicating that landlocked Alewife populations have specialized for life in freshwater and highlight the kidney as an important but understudied locus of osmoregulatory adaptation.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Leftovers from fish physiology in osmoregulation: a mechanism of the primary environmental cue inducing drinking in seawater, revisited

Sakamoto, Tatsuya[1] (*ryu@okayama-u.ac.jp*) Oral Presentation
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Abstract: For teleost fish in seawater, drinking the surrounding medium is essential to prevent dehydration. In mammals, salt taste can also trigger attractive behavioral responses; this pathway is selectively responsive to sodium, and the epithelial sodium channel (ENaC) is known to function as a component of the salt-taste-receptor system. In teleosts, however, chloride has been recognized as the primary environmental cue inducing drinking in seawater for more than half a century (Hirano 1974). Indeed, teleosts appear to have lost ENaC genes from their genomes.Against this background, we have identified a candidate molecule for chloride binding and chloride responsiveness in the medaka fish. Medaka genetically engineered to lack the corresponding gene exhibit a loss of behavioral attraction to seawater and impaired drinking responses. The ENaC-dependent salt-taste-receptor system is proposed to have evolved during terrestrial adaptation, coincident with the evolution of ENaC-activator “mineralocorticoid” aldosteronesignaling. Therefore, we may further discuss the evolution of the corticosteroid system. Specifically, we are identifying its ancestral form using structural biology as well in an acoel of the phylum Xenacoelomorpha, a lineage positioned near the base of deuterostome evolution. This presentation is also dedicated to the late Professor Tetsuya Hirano, our mentor.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Ontogeny of salinity tolerance and ionoregulatory plasticity in mummichog (Fundulus heteroclitus)

Berthelot, Evan[1] (*e.berthelot@unb.ca*) Oral Presentation
Co-Authors: Breves, Jason[2] and Zimmer, Alex[1]
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Abstract: Mummichogs (Fundulus heteroclitus) are an exceptionally euryhaline teleost that can tolerate the direct transfer between freshwater and seawater environments by compensating for the passive diffusion of ions via the plasticity of their ionoregulatory and osmoregulatory mechanisms, including the expression of ion-transporting proteins and ion-transporting cells, termed ionocytes. Although adult mummichogs possess a broad euryhaline capacity, previous studies have shown that early life stage mummichogs are less euryhaline, with the mechanisms underlying this discrepancy in euryhalinity remaining unclear. Thus, this research aimed to understand the relationship between ionoregulatory plasticity and salinity tolerance in early developmental stages of mummichog. We investigated changes in salinity tolerance by exposing early life stage mummichogs at various developmental stages to a range of salinities and monitoring survival. We found freshwater tolerance was limited at fertilization but increased with the onset of larval development, whereas brackish water and seawater tolerance persisted throughout development. We then assessed the plasticity of ionoregulatory mechanisms through gene expression and ion content analyses in both embryos and larvae. Prior to hatch, embryos showed few transcriptional responses to salinity transfer, whereas post-hatch larvae showed stronger responses to freshwater transfer. Embryos also had overall lower tissue ion concentrations in freshwater than in brackish or seawater, whereas larvae showed a greater capacity for maintaining ion balance across all salinities. Overall, salinity tolerance in developing mummichogs appears to be a function of reduced ionoregulatory plasticity, which may provide a physiological basis for ontogenetic changes in salinity tolerance in this species.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Functional classification of gill ionocytes in Pacific and Atlantic salmonids

Hiroi, Junya (*j-hiroi@marianna-u.ac.jp*) Oral Presentation
St. Marianna University School of Medecine

Abstract: To understand osmoregulation in salmonids, it is essential to establish a functional classification gill ionocytes, as has been well established in tilapia and zebrafish. In the present study, immunolocalization patterns of NKAα1a, NKAα1b, Rhcg1 and NBC1 were examined in the gills of Pacific salmonids (rainbow trout, masu salmon and chum salmon) and Atlantic salmon. In all species, gill ionocytes were classified into three types based on apical Rhcg1 and basolateral NBC1:

1) Rhcg1(+)/NBC1(-) ionocytes: These ionocytes were most common, basolateral NKAα1a was stronger than that of NKAα1b in freshwater-acclimated fishes and masu salmon parr, while NKAα1b became stronger in seawater-acclimated fishes and masu salmon smolt.
2) Rhcg1(-)/NBC1(+) ionocytes: These ionocytes were freshwater-specific, characterized by stronger NKAα1a.
3) Rhcg1(-)/NBC1(-) accessory cells: These cells were smaller in cell size, exhibited stronger NKAα1b immunoreactivity, which aids in distinguishing them from other ionocytes (which have weaker NKAα1b) in freshwater-acclimated fishes. In seawater-acclimated fishes, however, most ionocytes showed strong NKAα1b staining, making it difficult to distinguish accessory cells and other ionocytes with the current quadruple immunostaining approach (NKAα1a, NKAα1b, Rhcg1 and NBC1).
Salmonid Rhcg1(+)/NBC1(-) ionocytes appear homologous to tilapia type-III/IV ionocytes, while Rhcg1(-)/NBC1(+) ionocytes correspond to tilapia type-II ionocytes, despite salmonids lacking gill NCC2. We propose standardizing ionocyte nomenclature among teleosts.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

There and back again … almost: tales of an anadromous fish researcher

Shrimpton, Mark (*mark.shrimpton@unbc.ca*) Oral Presentation
University of Northern British Columbia

Abstract: Anadromous salmon are remarkable in the extensive migrations they undertake from freshwater to marine environments that maximize their reproductive potential. Not unlike our study organisms, as scientists we also travel to new environments, seeking to maximize our research potential. While studying at UBC, I found that environmental conditions affected the ability of juvenile Pacific salmon to successfully adapt to higher salinity. After my transcontinental “migration” to Massachusetts, we demonstrated that in Atlantic Salmon significant changes occur in the gill during the parr-smolt transformation before juveniles migrate into the ocean. Returning to the province of my academic home, work at UNBC demonstrated that physiological changes were also preparatory in adult Pacific salmon returning to freshwater. For some populations, there is amazing fidelity to specific spawning locations. The expectation is that philopatry will lead to local adaptation. However, we have recently demonstrated that straying to new locations may be a common pattern in anadromous salmon. Low levels of straying may be beneficial in providing temporal stability in population genetic structure. This is not unlike what I have encountered in my own career where working with colleagues from around the world has mutually benefitted our research and increased our understanding of the physiology of anadromous fishes.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Ready and swimming: ontogenetic changes preceding emigration in juvenile Alewife (Alosa pseudoharengus)

Schultz, Eric T.[1] (*eric.schultz@uconn.edu*) Oral Presentation
Co-Authors: Colby, Rebecca[2]; McCormick, Stephen D.[3]; Charmantier, Guy[4]; Goggins, Peter[1]; Li, Annchi[5]; Smith, Brandon P.[6]; Stephens, Jacqueline B.[7] and Sternberg, Kayla M.[8]
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Abstract: Diadromous fishes complete salinity transitions along their migratory routes, requiring robust physiological mechanisms for survival. While some fishes such as salmonids undergo conspicuous external modifications and well-known physiological changes prior to migration, others such as Alewife do not show obvious external changes and internal preparatory changes have not been reported. We tested juvenile alewives in two ontogenetic stages, pre-migrant and migrant, for shifts in physiological processes, morphometry, physical state, and swimming performance associated with increased migratory fitness. We exposed subjects to freshwater (0.5 ppt) or seawater (35 ppt) for 1 or 5 days and detected no differences between stages in the activity of gill Na+ K+-ATPase. Migrants were unexpectedly more likely to die over 5 days than pre-migrants when exposed to seawater. In comparison with pre-migrants, migrants are larger, older, have more fusiform bodies, and have greater ability to osmoregulate and swim in seawater. These results indicate that Alewife undergo preparation for outmigration to support fitness and survival in the marine environment. Increasing our understanding of how juveniles interact with their environment during migration is crucial for current-day management of diadromous species, as well as for illuminating the evolutionary history of lifestyles involving salinity transitions.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

The genomic underpinnings of adaptation to freshwater in multiple, independently derived populations of landlocked alewife

Velotta, Jonathan[1] (*Jonathan.Velotta@du.edu*) Oral Presentation
Co-Author: Corcoran, Riley[1]; Schultz, Eric T.[2]
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Abstract: In the northeastern coastal USA, anadromous alewives have been repeatedly and independently trapped in freshwater above dams since European colonization. This system provides a unique natural evolutionary experiment to test one of the fundamental questions of Steve McCormick's pioneering career: what physiological changes accompany life history changes? In this study, we extend to the genomic level work that Steve helped build. We have shown previously that freshwater or "landlocked" populations of Alewife (Alosa pseudoharengus) have repeatedly evolved reduced tolerance of and performance in seawater. Here, we compared whole genomes of four landlocked and one

anadromous population to test whether changes at the genomic level show comparably repeatable changes upon landlocking. We found that repeated positive selection at the genomic level is rare, limited to a single region on a single chromosome. Despite this, a targeted candidate gene analysis revealed that regions of selection do occur in genes with function in osmoregulation in some populations though not all. Surprisingly, the strongest signal of selection in the genome was not one of positive selection, but one of conserved, balancing selection in a single gene family known as protocadherins, which play a role in neural circuit formation and neuron recognition. Our results suggest that repeated life history changes in alewives are driven by directional changes to osmoregulatory physiology and by the maintenance of genetic diversity in nervous system functions that may improve navigation in lake habitats. In keeping with Steve's legacy, our results provide fundamental insight into the evolutionary processes that shape the physiology of anadromous fishes.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

An increased thyroid hormone apparatus in the brain of chum salmon during smoltification

Wong, Marty Kwok-Shing[1] (*martywong@aori.u-tokyo.ac.jp*) Oral Presentation
Co-Authors: McCormick, Stephen D.[2]; Tsukada, Takehiro[3]; Hyodo, Susumu[1]
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Abstract: Chum salmon (Oncorhynchus keta) migrates to the sea during early fingerling stage, a much earlier stage than for most salmonids. The smoltification process and control in chum salmon are unclear, probably due to their small size and inconspicuous morphological changes.We demonstrated that chum salmon alevins increase their salinity tolerance sharply at the stage of emergence from gravels coinciding with the upregulations of Na/K-ATPase alpha 1b (NKA a1b),Na–K–Cl cotransporter 1 (NKCC1)andcystic fibrosis transmembrane conductance regulator (CFTR), which favor an ion-excreting function in seawater.Thus, the emergence from gravels in chum salmon coincides with the smoltification window, which occurs later in other salmonids. We found an increase in brain deiodinase 2 (DIO2) during smoltification in FW and a decrease in deiodinase 3 (DIO3) were triggered by seawater exposure, indicating a local increase of thyroid hormone in the brain.A general upregulation of thyroid hormone receptor alpha-B (THRB) in the brain during emergence also supports the idea that thyroid hormone action is increased. The trigger for smoltification in chum salmon is likely integrated into their embryonic development and external stimuli such as photoperiods may have minimal roles. The consistent size and stages of chum salmon, along with the sharp smoltification window, make it an excellent comparative model to elucidate the endocrine regulation of smoltification in salmonids.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Transcriptomic remodeling of gill ion transport and respiratory mechanisms during metamorphosis in sea lamprey

Shaughnessy, Ciaran[1] (*ciaran.shaughnessy@okstate.edu*) Oral Presentation
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Abstract: Metamorphosis in the sea lamprey (Petromyzon marinus) includes the transformation of freshwater-resident larvae into juveniles capable of surviving in seawater. To more broadly identify functional genomic changes underlying this shift, we analyzed RNAseq datasets from gill tissue (including red blood cells) comparing larvae and juveniles, focusing on genes associated with ion transport, acid-base regulation, respiration, epithelial barrier function and remodeling, and endocrine signaling. Several ion transport pathways associated with the seawater-type ionocyte model of ion secretion were strongly upregulated following metamorphosis, including isoform-specific transcripts encoding subunits of the Na+/K+-ATPase (nka), Na+-K+-2Cl- cotransporter (nkcc1), anoctamin 1 (ano1), inwardly rectifying K+ channel (kir4), and NH4+ transporters (rhcg). Metamorphosis was also associated with remodeling of epithelial barrier components, with a large number of claudins (cldn) expressed in the gill and multiple isoforms showing strong upregulation. Evidence of physiological reprogramming was also apparent in shifts among isoforms of key metabolic and respiratory genes, including the previously described phenomenon of isoform switching among carbonic anhydrases (ca). Interestingly, transcription of globin (glob) genes displayed marked shifts in dominant isoforms, with some larval isoforms disappearing and other isoforms appearing in juveniles. Several endocrine and developmental regulatory pathways appeared to be associated with metamorphosis, including expression of progestin receptor (pr), somatostatin receptor (sstr), insulin-like growth factor receptor (igfr) and binding proteins (igfbp), and thyroid hormone receptor (thr) and interacting protein (trip). Together, these results highlight coordinated transcriptional reprogramming of ion transport, epithelial structure, and regulatory pathways during preparation for the freshwater-to-marine transition of sea lamprey.

IONS II Environmental physiology of life-history transitions: A tribute to Steve McCormick

Paternal effects of diethyl phthalate (DEP) and cadmium (Cd) on the embryonic development of its offspring

Wu, Su Mei (*sumei@mail.ncyu.edu.tw*) Poster Presentation
National Chiayi University

Abstract: Our previous studies had found that some endocrine- disrupting chemicals, such as DEP (diethyl phthalate) and cadmium (Cd2+), significantly induced the levels of 17ß-esterdiol (E2) in female zebrafish and through to their larvae through maternal effects, consequently inducing abnormal development of cartilage in the pharyngeal arches of

their embryos. However, the effects from the paternal side were not clear. In addition, post studies had found that paternal BPA exposure alters craniofacial cartilage development, leading to abnormalities in its offspring. Therefore, we hypothesized that paternal DEP and Cd will also induce some irregularities on its offspring because both chemicals can induce upregulation of E2 levels in exposed males. The present study investigates the impacts of paternal effects upon male zebrafish exposure to 0, 4.45, 8.9, and 17.8 μM of Cd2+ and 0, 8.06, 16.12 μM of DEP for 5 days. We found the Cd2+ contents showed a dose response in the testis, and the mt2 mRNA of the liver was significantly increased in males exposed to 17.8 μM of Cd. However, some genes related to testis quality (bcl2a, dmrt1, and baxa), methylation (dnmt3 and tekt2), E2 inducing (cyp19a1a), and all mRNA expression were not significantly different after the male zebrafish exposure to neither DEP nor Cd for 5 days. But their embryos showed abnormal craniofacial development. We are interesting to know the reasons in future studies.

TROPICAL Tropical fish: A tribute to Dave Randall

Tropical fish: A tribute to Dave Randall

Adalberto Val and Vera Almeida-Val

Abstract: This symposium will celebrate the legacy of Dr. David Randall, whose groundbreaking contributions to fish physiology, especially in tropical systems, have profoundly influenced generations of researchers. It aims to honor his memory by presenting the latest findings on tropical fish from across the globe, showcasing new research frontiers and emerging technologies that are deepening our understanding of these species and their environments. From physiological plasticity to environmental adaptation, the symposium will highlight how Dr. Randall's vision continues to inspire cutting-edge science in tropical ichthyology. We warmly welcome all who share a passion for tropical fish, as Dave did, and a commitment to understanding the environmental challenges shaping their future.

TROPICAL Tropical fish: A tribute to Dave Randall

Life at the Edge: David Randall's Alpha Helix Legacy in the Amazon

Val, Adalberto (*dalval.inpa@gmail.com*) Oral Presentation
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Abstract: In 1976, exactly fifty years ago, the research vessel Alpha Helix anchored in the floodplain lakes of the central Amazon, turning one of Earth's most remote waters into a laboratory at the edge of life. There, David Randall and colleagues confronted a fundamental biological question: how can fishes breathe, function, and persist where oxygen is scarce, waters are warm, and environmental stability is only an illusion? Their work revealed that species such as Arapaima gigas survive by reorganizing respiratory function, relying on a lung-like swim bladder for aerial oxygen uptake while maintaining gill pathways essential for carbon dioxide elimination, redefining vertebrate gas exchange across water and air. At the cellular level, the discovery of large pools of inositol pentaphosphate in erythrocytes exposed a molecular mechanism capable of fine-tuning hemoglobin–oxygen affinity, providing a biochemical foundation for life under chronic hypoxia. These discoveries established the Amazon as a proving ground for integrative physiology, linking environment, organism, and molecule across levels of biological organization. Contemporary research now reveals the genetic and biochemical regulation of inositol phosphate metabolism underlying oxygen transport in air-breathing fishes, extending the mechanistic framework first illuminated aboard the Dave Randall's Alpha Helix expedition. Yet the conditions that shaped these adaptations are rapidly changing. Warming waters, intensifying droughts, and declining oxygen availability are compressing physiological safety margins, exposing limits of evolutionary specialization, and redefining the capacity of Amazonian fishes to persist in a rapidly changing world. (INCT ADAPTA III, CNPq, CAPES).

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The tambaqui (Colossoma macropomum), a model neotropical species in fish physiology and toxicology

Wood, Chris[1] (*woodcm@zoology.ubc.ca*) Oral Presentation
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Abstract: With his leadership of the Alpha Helix Expedition of 1976 and his many contributions thereafter, Dave Randall brought the mechanistic approach to fish physiology in Brazil; his legacy continues today. In the northern hemisphere, Dave developed the rainbow trout as a model cold-water species for mechanistic research. The Characiform tambaqui now plays a comparable role as a model warm-water neotropical species for mechanistic research. Native to both acidic black-water and the circum-neutral white-water of the Orinoco and Amazon watersheds, this species is now widely available through aquaculture on three continents. Its genome has been sequenced. We will highlight some of the key findings over the past 30 years of our large collaborative team on the mechanisms underlying its nitrogen metabolism, osmoregulation, respiratory gas transport, acid-base regulation, and responses to low and high pH, metals, dissolved organic carbon (DOC), feeding, exercise, hypoxia, and climate change. (INCT ADAPTA I, II, & III – CNPq; NSERC).

TROPICAL Tropical fish: A tribute to Dave Randall

Variability and Diversity: Key Factors in Freshwater Fish Adaptation

Almeida-Val, Vera Maria Fonseca de (*veraval30@gmail.com*) Oral Presentation
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Abstract: Amazon Basin resulted from diverse tectonic processes and reached its current form after the uplift of the Andes. Notably, the evolutionary history of some fish species of the Amazon predates the basin itself. It is formed by a heterogeneous environment that connects rivers with distinct physicochemical characteristics. Some rivers experienced course shifts due to tectonic movements, leading to river captures, the formation of waterfalls, and temporary lakes. To survive these changes, fish evolved diverse adaptive strategies. Phenotypic and genotypic plasticity of Amazonian fish have been documented in our lab over the past 45 years, explaining their ability to adapt to environments with little or no oxygen, variable temperatures, and acidic waters. Herein, we present examples of adaptation: genetic variability in populations inhabiting different environments; and phenotypic plasticity observed in populations inhabiting different environments. The distribution of a species may consist of contiguous populations, as seen in the red-bellied piranha, which shows greater genetic differentiation with increasing geographic distance. Distant populations diverge genetically because they no longer interbreed, demonstrating locally unique genetic variation associated with adaptation. Phenotypic plasticity is found in populations of a species occurring in different rivers. Distinct patterns of gene

expression occur in genes related to osmoregulation, which are associated with specific water types (black, clear, or white rivers). A third mechanism of adaptability operates over longer evolutionary timescales, i.e. selective pressure on critical genes for fish survival that show positive selection; lactate dehydrogenase (LDH) is a well-documented example. (INCT ADAPTA III, CNPq, CAPES).

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Current knowledge of lunar synchronized spawning in fish

Fukunaga, Kodai (*h217161@cs.u-ryukyu.ac.jp*) Oral Presentation
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Abstract: Many organisms are sensitive to lunar cycle-related changes, such as tidal fluctuations and variations in moonlight intensity. Marine organisms must synchronize their behavior to the underwater environment, which is more complex and variable than terrestrial habitats. Lunar-synchronized spawning refers to the monthly spawning cycle triggered by changes in phase of the moon and is especially common among temperate and tropical marine species. Corals, marine worms, and fish are well-known examples of organisms that spawn synchronously with lunar phases to improve fertilization success. Research over the past decade has provided insights into lunar-synchronized spawning, and studies of families such as Siganidae and Serranidae have begun to elucidate the underlying mechanisms in fish. Field investigations, particularly in tropical and subtropical waters, have shown that many species spawn during periods of the new or full moon. Species-specific spawning times may be determined genetically, with long-term environmental adaptation influenced by physicochemical and ecological factors. Even within the same species, spawning moon phases can vary by location. As lunar-synchronized spawning is regulated by moonlight cues, these geographical differences are likely driven by genetic variation in photoresponsiveness. Physiological experiments have further suggested that nocturnal light plays a role in both gonadal development and the timing of spawning in fish. Fish have been hypothesized to be able to predict moon phases for spawning by detecting temporal changes in moonlight intensity associated with the lunar cycle.

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External factors influencing air breathing behaviour of a facultative, gut air breathing fish, Panaque maccus

Nelson, Jay (*jnelson@towson.edu*) Oral Presentation
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Abstract: The Neotropics are home to the richest freshwater fish fauna in the world, including over 1000 species of Loricariidae many of whom can use their gut as an air

breathing organ (GAB fish). Some air breathing fish species have been studied extensively, yet little is known about the many gut air breathers and what dictates when they breath air. Gut air breathing presumably evolved to cope with hypoxic waters, but it also introduced new predator regimes and evolutionary success may have necessitated co-evolution of avoidance strategies. In addition, conspecific dominance hierarchies often exist in fish communities and these may influence an individual's inclination to breath air. Finally, there is little understanding of how GAB fishes balance the digestive and respiratory functions of their gut and whether eating influences air-breathing behaviour. The purpose of this study was to investigate the impact of perceived predation threat, position in a dominance hierarchy and diet on the air-breathing behaviour of the facultative air-breathingPanaque maccus. Air breathing frequency under hypoxia was a highly repeatable measurement among individuals and was deterred by the presence of a predaceous bird decoy, especially if the decoy was in a strike position. Groups ofP. maccusheld together established dominance hierarchies amongst themselves and air breathing frequency and onset were significant functions of position within a dominance hierarchy. Fed and starvedP. maccusexhibited no difference in air breathing behaviour under hypoxia however there was some indication that a more digestible diet (less fiber) required more air-breathing under hypoxia.

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Tracking tetra transport: how do conditions change for a popular ornamental fish in Brazil?

Addison Marcos, Alexander[1] (*alexander.marcos@uws.ac.uk*) Oral Presentation
Co-Authors: Kochhann, Daiani[2]; Snellgrove, Donna[3]; Val, Adalberto[4]; Killen, Shaun[5]; Azambuja, Guilherme[4]; Alexander, Mhairi[1] and Sloman, Katherine[1]
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Abstract: The cardinal tetra (Paracheirodon axelrodi) is one of the most recognisable and popular fish in the ornamental trade. Originating from the Rio Orinoco and Rio Negro in the Brazilian Amazon, many Indigenous communities have relied on the trade of this and other ornamental species for decades. Despite its popularity, little is known about the conditions these fish experience on their journey from their point of capture until they arrive in a home aquarium. The water quality fish are exposed to changes during transport where deterioration can impact fish physiology, behaviour and ultimately welfare. To improve our understanding of the conditions cardinal tetra experience, water samples were taken at various stages of the journey from local fishing communities in the Amazon to arrival at holding facilities in Manaus. Fish were also assessed for condition, colour and behaviour throughout the journey. Water quality varied depending on the stream where fish were captured, and they experienced significant changes in water quality, particularly during the boat journey to Manaus. Results from this study will help inform further research on ornamental fish transport and provide much needed knowledge about an under-researched fishery of a very well-known ornamental fish.

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Microplastic contamination in fish digestive tracts from the Rio Negro and Rio Solimões during an extreme low-water period

Tuvikene, Arvo (*arvo.tuvikene@emu.ee*) Oral Presentation
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Abstract: Microplastics (MPs) are widely detected in the gastrointestinal tracts of aquatic organisms. We assessed plastic contamination in two Amazonian river systems, Rio Negro and Rio Solimões, examining both the aquatic environment and fish digestive tracts. Sampling was conducted in November–December 2023 during the low-water season of an exceptionally dry year. We analysed MP abundance, size (0.04–5.0 mm), shape, and polymer type, and compared contamination among fish species representing different feeding guilds (piscivorous, planktivorous, herbivorous, invertivorous, detritivorous, omnivorous). In total, 128 individuals were examined; MPs were detected in 84.0% of fish from the Prato Canal and 87.4% from Vila do Janauacá. Mean MP loads were 3.3 ± 2.9 particles per fish (Prato Canal) and 2.8 ± 2.1 (Vila do Janauacá). No significant relationships were found between fish length or mass and MP abundance or particle size. MP size differed significantly between river systems and was reflected in fish: individuals from the Solimões contained larger particles on average than those from the Rio Negro. Feeding guild patterns varied between sites: piscivorous fish had the highest MP loads at Vila do Janauacá, whereas planktivorous fish were most contaminated in the Prato Canal. Most ingested MPs were filaments, followed by fragments. The dominant polymers were polyamide (nylon), polyethylene terephthalate, and polypropylene. These results demonstrate substantial MP contamination even in relatively remote Amazonian environments. Increased rainfall and higher water levels may enhance MP transport and availability, potentially leading to greater accumulation in fish, particularly in visually oriented foragers.

TROPICAL Tropical fish: A tribute to Dave Randall

Altered temperature and pH modify key biomarker responses to gasoline exposure in a Neotropical fish

Dal Pont, Giorgi[1] (*giorgidalpont@gmail.com*) Oral Presentation
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Abstract: We investigated how thermal (30 ºC) and acid (pH 4.0) stress modulate the biochemical responses of the Neotropical fish Astyanax altiparanae to the water-soluble fraction of gasoline (WSFG). Chemical analysis revealed that thermal stress caused a complete loss of volatile low molecular weight BTEX and PAHs compounds, fundamentally

altering the exposure profile and influencing biotransformation enzymes. Specifically, while WSFG induced hepatic ethoxyresorufin-O-deethylase (EROD) activity under control conditions, thermal stress inhibited this response, and acid stress nullified it. Oxidative stress, measured as protein carbonyl content, was affected by isolated thermal and acid conditions and was significantly exacerbated when acid stress was combined with WSFG exposure. This suggests the antioxidant defense system was insufficient to prevent cellular damage, as supported by the lack of a catalase (CAT) response across treatments. Furthermore, neurotoxic responses varied significantly; brain acetylcholinesterase (AChE) activity was up-regulated by thermal stress but inhibited by acid stress. These findings demonstrate that environmental stressors alter both contaminant bioavailability and the resulting biomarker responses. Consequently, the interpretive value of these biomarkers may be reduced in multi-stressor scenarios, suggesting they should be used with care rather than as definitive indicators. This study indicates that biomarker patterns are highly dependent on prevailing conditions, which may complicate their application in environmental monitoring.

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Climate-change stress reshapes mRNA-miRNA regulatory networks in the Amazonian fish Apistogramma agassizii

Heinrichs-Caldas, Waldir (*dinhein@gmail.com*) Oral Presentation
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Abstract: The accelerating impacts of climate change pose serious risks to animals and ecosystems. Projected increases in atmospheric CO_2 and temperature are already limiting for some species, triggering negative physiological, biochemical, and genetic responses. Gene expression can be modulated by epigenetic mechanisms, including microRNAs (miRNAs), non-coding RNAs that regulate messenger RNAs (mRNAs) through post-transcriptional control. Despite advances in epigenetics, studies linking fish epigenetic responses to climate-change stressors remain scarce, limiting our understanding of miRNA function under warming and elevated CO_2. Here, we investigated the role of miRNAs in shaping gene-expression responses to an extreme climate-change scenario proposed by the IPCC and assessed post-transcriptional regulatory effects using in silico analyses. Fish were exposed to conditions simulating this extreme scenario, and transcriptomic profiling revealed 968 differentially expressed genes (533 upregulated and 435 downregulated), of which 67% received functional annotation. Upregulated genes were mainly associated with membrane transport, channel and transporter activity, and plasma-membrane functions. In contrast, downregulated genes were enriched for central metabolic pathways, lipid and steroid metabolism, and energy-related processes. Small RNA sequencing identified 255 differentially expressed miRNAs, with 11 showing significant differential expression. Integrative mRNA–miRNA analysis yielded 518 putative interaction pairs: increased expression of miR-206-3p and miR-724-5p was associated with multiple downregulated

mRNAs, whereas reduced miR-101b-5p expression was linked to increased mRNA levels. Together, these findings suggest that miRNAs contribute to post-transcriptional regulation of key metabolic and membrane-related pathways in A. agassizii under extreme climate-change conditions.

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Developing thermal death time models from CTmax data to assess thermal tolerance in Colossoma macropomum

Moura, Lucas[1] (*moura@unbc.ca*) Oral Presentation
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Abstract: The unprecedented heat waves observed in Amazon freshwaters in recent years underscore the urgent need to move beyond metrics of relative thermal tolerance toward predictive models of temperature-driven mortality in fishes. Currently, quantifying thermal mortality at the population level is challenging because critical thermal maximum (CTmax) estimates do not capture the time dependency of thermal stress. This limitation can be addressed using recently developed approaches that convert dynamic CTmax values, derived at different ramping rates and/or starting temperatures, into parameters of the thermal death time (TDT) model. The TDT model formalizes the relationship between temperature and time to mortality using two parameters: static CTmax1h (the temperature causing mortality within 1 hour) and thermal sensitivity (z), defined as the temperature change resulting in an order-of-magnitude shift in time to mortality. In this study, we developed a TDT model for juvenile tambaqui (Colossoma macropomum) using data from dynamic CTmax trials. Fish were acclimated to 27, 30, or 33°C for 15 days and subjected to three warming ramping rates (0.2, 0.3, or 0.4°C min^{-1}) under normoxic and hypoxic (<2 mg L^{-1}) conditions until loss of equilibrium. Static CTmax1h ranged from 38.0 to 41.1°C across acclimation temperatures, whereas hypoxia had minimal impact on static CTmax1h. However, hypoxia significantly reduced thermal sensitivity, resulting in shorter survival times at smaller temperature increases than under normoxia. Ongoing work aims to integrate TDT models with field-temperature data to quantify survival under realistic thermal scenarios, providing a mechanistic basis for predicting mortality during heatwaves. (INCT ADAPTA III – CNPq, CAPES; NSERC).

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Behavioural and physiological selection in a tropical ornamental fishery: Evidence from Amazonian species

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Abstract: Understanding the behavioural and physiological traits of wild fish populations is essential for predicting responses to anthropogenic and environmental pressures. Wild harvest represents one such pressure, with important implications for selection and evolution. While this has been widely examined in commercial fisheries, it remains largely unexplored in smaller-scale artisanal fisheries, which typically occur in tropical systems where high biodiversity and environmental variability shape trait expression. Here, we combine laboratory and field-based approaches to investigate the behavioural, physiological, and morphological traits that determine capture vulnerability, and assess the potential for gear-specific selection in a popular tropical ornamental species, the cardinal tetra (Paracheirodon axelrodi). In the lab, individuals with a higher capture vulnerability were smaller, more exploratory, and fatigued more rapidly. Comparisons of cardinal tetra captured with passive traps versus active nets in the field also revealed distinct phenotypic differences: trap-caught individuals were more exploratory, whereas net-caught fish were more cohesive, and exhibited stronger swimming performance. Field-based underwater video recordings further documented complex behaviours during the capture process, which differed between species and environmental conditions. Collectively, these findings provide important insight into trait-based and gear-specific selection in the Amazonian ornamental trade.

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Physiological plasticity of tropical coral reef fishes during an extreme marine heatwave on the Great Barrier Reef

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James Cook University

Abstract: Tropical coral reef fishes routinely experience high and variable temperatures; however, the scope of their physiological plasticity under extreme marine heatwaves remains poorly resolved. Leveraging the 2016 mass bleaching event on the Great Barrier Reef (GBR, Australia) as a natural experiment, we quantified integrated responses in nine common reef fish species (Families Pomacentridae and Apogonidae) collected from reefs around Lizard Island across four time points: prior to heat stress, early warming, peak temperatures, and post-heatwave recovery. Building on our previously published liver transcriptomic analyses from this event, which revealed species-specific yet functionally overlapping molecular responses to prolonged thermal exposure, we examined whether

these gene expression shifts were reflected in organismal phenotype. Across species, peak warming was associated with altered body condition and gut fullness consistent with changes in energetic balance, remodelling of gill morphology, and shifts in metabolic enzyme activities in gill and muscle tissues (citrate synthase and lactate dehydrogenase). These patterns indicate restructuring of aerobic and anaerobic capacity during acute thermal exposure. However, several physiological metrics showed incomplete recovery following temperature decline, suggesting that short-term plasticity may incur sustained energetic costs. Together, these findings demonstrate that marine heatwaves drive coordinated molecular, biochemical, and structural adjustments in wild tropical reef fishes. By linking transcriptomic responses with tissue-level metabolism and whole-animal condition across a real climate extreme, this work extends classic comparative frameworks of tropical fish physiology into the context of increasingly frequent marine heatwaves and against the backdrop of ongoing ocean warming.

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It's O"K+"- a role for potassium in ion regulation for fish living in the acidic waters of the Rio Negro.

Goss, Greg[1] *(ggoss@ualberta.ca*) Oral Presentation
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Abstract: Acidic, low-ionic-strength freshwater environments, such as the Rio Negro ([Na+] ~ 10 uM, [K+] ~ 4 uM, [Ca2+] ~20 uM, pH 3.5-4.5), pose a thermodynamic challenge to the function of electroneutral Na+/H+ exchangers (NHE). Recently, Clifford et al. (2022) described a K+-efflux-dependent Na+ uptake (JinNa+) mechanism in zebrafish to overcome these challenges. Using radiotracer (22Na+) techniques, we investigated the ionoregulatory mechanisms of tambaqui (Colossoma macropomum, ~15-35 g) under mild (pH ~6.0) and highly acidic (pH 3.8) conditions. Acute exposure to pH 3.8 resulted in an initial ~30% reduction in JinNa+ after 2 h of pH 3.8 exposure when compared to control (pH 6.0). However, after 18 h in pH 3.8, JinNa+ was increased by~110% over control values. Notably, there was no increase in [K+] in the water, unlike that found in zebrafish. However., graded addition of [KCl] (from 0.125 mM- 10 mM) to the water resulted in concentration-dependent reductions in JinNa+, with > 95% reduction at 10 mM [KCl]. Similar reductions in JinNa+ were also seen with a graded NH4Cl. JinNa+ was not dependent on alterations of TEP caused by either 10 mM KCl or NH4Cl since 10 mM CaCl2 resulted in a similar change in TEP without any effect on JinNa+. At low [K+], an inwardly directed net K+ gain was noted and revealed a high-affinity inwardly-directed K+ transport was occurring. We suggest an overlooked role for K+ in mediating Na+ uptake in low pH, low ionic strength waters. (INCT ADAPTA III – CNPq; NSERC).

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The air breathing physiology and temperature and hypoxia tolerance of the oriental weatherloach (Misgurnus anguillicautatus)

Burggren, Warren (*burggren@unt.edu*) Oral Presentation
Co-Authors: Burbules, Katherine
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Abstract: Air breathing has evolved in multiple fish species as an adaptation to hypoxia and other extreme conditions. The air-breathing weather loach, Misgurnus anguilicaudatus, has a modified hindgutthat facilitates oxygen uptake through the intestinal walls of the swallowed air bubble. We hypothesized that air breathing would facilitate recovery from metabolic stress resulting from elevated temperatures and/or hypoxia.To investigate the aerial and aquatic contributions to gas exchange during acute changes in aquatic PO2 and temperature, we employed a closed respirometry system that measured combined aquatic and atmospheric oxygen consumption ($\dot{M}O2$) as well as gill and airbreathing ventilation frequency in unrestrained, undisturbed loaches as well as during enforced activity. When exposed to increasing degrees of hypoxia (21 kPa to 5 kPa) at 23◦C, air breathing frequency increased (~6 bph to 100+ bph). However, gill ventilation frequency initially increased then dropped as hypoxia intensified. Critical partial pressures for maintaining $\dot{M}O2$ (PCrit) increased as temperature increased, as expected, but then decreased again between 27◦C and 31◦Ck. Overall, weather loaches showed faster recovery from stress (enforced activity) and increased temperature and hypoxia tolerances compared to non-air-breathing fish. The respiratory response of subtropical air-breathing fish under seasonal change scenario in natural populations follows unexpected patterns indicating the possibility of unknown physiological influences and a significant effect of air-breathing on metabolism.

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Rapid transformation in respiratory performance of coral reef fish larvae settling on a reef

Nilsson, Göran *g.*(*e.nilsson@ibv.uio.no*) Oral Presentation
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Abstract: In relation to body size the fastest swimming fish are probably larvae of coral-reef fish on their way to settle on reefs. Swimming speeds up to 60 body-lengths per second have been recorded in these one-centimeter-long fishes. The high swimming speeds of the pre-settlement larvae are accompanied by the highest rates of oxygen uptake ever recorded in ectothermic vertebrates (over 5 000 mg oxygen per kg and hour). These high rates of oxygen uptake come at the cost of poor hypoxia tolerance, and hypoxia tolerance is needed for a life among coral branches, where oxygen levels may fall below 20 % of air saturation at night. Swim-respirometry shows that at the time the larvae settle on the reef they go through a striking respiratory transformation. The maximum rate of oxygen uptake falls, while ability for high-affinity oxygen uptake at low oxygen levels is increased.

Mechanistically, this transition in respiratory performance probably involves a change in the expression of hemoglobin isoforms, from low to high oxygen affinity.

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Aerial and aquatic oxygen uptake during recovery from exercise and air exposure in Arapaima gigas

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Abstract: The pirarucu Arapaima gigas is an obligate air-breathing fish with a massive internal air-breathing organ yet also retains substantial gills. In this study, Arapaima was exercised for 10 minutes either in water or in air, or it was exposed to air for one hour. During subsequent recovery from exercise or air exposure aerial and aquatic oxygen uptake and ventilatory activity were recorded for two hours. Compared to the initial control period MO2 was almost doubled during air exposure. A doubling of total MO2 was also recorded during the first 30 minutes of recovery in all three experimental series. Thereafter MO2 returned to resting values, indicating a quick recovery from exercise as well as from air exposure. Aerial oxygen uptake contributed between 70% and 80% to total MO2, and this did not change during recovery from the different treatments. Aerial and aquatic ventilation was significantly elevated during initial recovery from all treatments. Ventilatory activity returned to resting values more quickly after exercise in water or air exposure, than after exercise in air. Relating oxygen uptake from air and water to ventilatory activity revealed that breathing air was about 200-fold more effective than breathing water. Nevertheless, the elevated oxygen consumption during early recovery did not enhance aerial respiration more than aquatic respiration, irrespective of total MO2 aerial respiration contributed between 70% and 80% to total MO2. (NSERC, INCT ADAPTA III - CNPq)

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Chronic climate stress drives gill remodeling and energetic demand in Amazonian fish

Braz-Mota, Susana (*susanabmota@gmail.com*) Oral Presentation
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Abstract: Climate change is expected to intensify global warming and increase CO2 concentrations, posing particular risks to Amazonian fish that already live near their upper thermal limits. We investigated the effects of long-term exposure (550 days) to an extreme 2100 climate scenario (+4.5 °C and ~1020 ppm CO2) on the tambaqui (Colossoma macropomum). Fish were maintained under controlled present-day and future conditions, and we assessed mitochondrial respiration in permeabilized gill tissue, ionoregulatory enzyme activity, and gill morphology. Chronic exposure increased carbonic anhydrase, Na^+/K^+-ATPase, and H^+-ATPase activities, indicating enhanced acid–base regulation and

ion transport, and suggesting higher energetic demands at the gill level. Mitochondrial respiration also increased under future conditions. Overall, long-term exposure to projected climate extremes may shift energy allocation toward maintenance processes, potentially compromising performance and threatening the persistence of this native Amazonian species.

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Overcoming DNA degradation in passive eDNA sampling for ichthyofauna monitoring through optimized filter selection and short-fragment markers

Dal Pont, Giorgi[1] (*giorgidalpont@gmail.com*) Oral Presentation
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Abstract: Environmental DNA (eDNA) has emerged as a transformative tool for aquatic biodiversity assessment, with a notable impact on ichthyofauna monitoring. However, the logistical demands of active filtration often limit its broad-scale application. Passive filtration represents a critical advancement for the field, offering a low-cost, operationally simplified alternative that facilitates longitudinal monitoring in remote or resource-limited environments. This study evaluated the efficiency of five distinct filter media [mixed cellulose esters (0.45 µm), glass fiber (0.8 µm), cellulose nitrate (0.45 µm), nylon (0.45 µm), and polyethersulfone (0.45 µm)] for eDNA retention via 24-h passive exposure at the Marine Aquarium of Paranaguá, Brazil. Methodological processing involved DNA preservation in 99% ethanol, extraction using the Blood and Tissue kit, Qubit fluorometric quantification, and NanoDrop purity assessment. Significant variance in performance was observed (ANOVA, $p<0.001$), with Tukey's post-hoc analysis identifying nylon filters as the superior substrate, yielding significantly higher DNA concentrations (mean ~3.5 ng/µL; $p<0.01$) and the highest amplification success. Molecular detection via PCR targeted the COI and 12S (MiFish, NeoFish, and Teleo) markers. While the COI marker failed to amplify, likely due to the fragmented nature of environmental templates, the 12S Teleo marker successfully detected eDNA across multiple samples. A Tilapia DNA spike-in assay confirmed the absence of PCR inhibition, attributing the COI failure to the accumulation of degraded genetic material during the 24-h exposure period. These results demonstrate that the integration of nylon filters with sensitive, short-fragment markers provides an optimized framework for non-invasive ichthyofauna monitoring in anthropogenically impacted ecosystems.

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Validation of environmental DNA metabarcoding for seasonal fish monitoring in the São Salvador reservoir: Identifying hidden biodiversity in the Tocantins River, Brazil

Dal Pont, Giorgi[1] (*giorgidalpont@gmail.com*) Oral Presentation
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Abstract: This study evaluates the efficacy of environmental DNA (eDNA) metabarcoding for monitoring fish biodiversity in the UHE São Salvador reservoir, Tocantins River, Brazil. Four eDNA sampling campaigns were conducted between 2022 and 2023, encompassing two separate collections during the dry season and two during the rainy season to account for seasonal variations. eDNA water samples were used to identify fish species presence, followed by a comparative analysis between the generated DNA sequence data (reads) and historical records obtained through traditional physical sampling methods. Comparative analysis between metabarcoding data and historical records revealed that the number of reads significantly correlates with species abundance. These findings align with classic ecological patterns, characterized by a few highly dominant species and a high frequency of rare taxa. Notably, the eDNA approach identified 18 species previously unrecorded in traditional surveys, demonstrating its superior sensitivity in detecting organisms with low abundance or cryptic behaviors that often evade physical capture. While metabarcoding is subject to limitations such as DNA amplification biases and varying sampling efforts, it remains a robust and promising tool for modern environmental assessments. By providing a more granular view of community dynamics and identifying previously overlooked biodiversity, this method significantly enhances the capacity to develop targeted conservation and management strategies for anthropogenically impacted aquatic ecosystems. Ultimately, these results validate metabarcoding as a promising instrument for longitudinal monitoring, offering critical insights into how ichthyofauna populations adjust to habitat alterations and human pressures in large reservoirs.

TROPICAL Tropical fish: A tribute to Dave Randall

“Is there reversibility in the expression of microRNAs in Colossoma macropomum exposed to an extreme climate change scenario?”

Sanches, Carlos Eduardo da Silva (*carloseduardosilvasanches@gmail.com*) Oral Presentation
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INPA - Instituto Nacional de Pesquisas da Amazônia

Abstract: MicroRNAs (miRNAs) are conserved regulators of gene expression in fish and may contribute to transgenerational responses to environmental change. Climate change–related stressors, including elevated temperature and CO_2 and reduced dissolved oxygen, induce oxidative stress and trigger metabolic, immunological, endocrine, and behavioral

adjustments. In the brain, these responses often involve enhanced anaerobic metabolism, strengthened antioxidant defenses, and cortisol-mediated mechanisms that sustain glucose availability. Such physiological shifts may be accompanied by epigenetic regulation. Epigenetics comprises heritable changes in gene expression without alterations in DNA sequences, mediated by mechanisms such as DNA methylation, chromatin remodeling, and RNA interference. Within this framework, miRNAs are small non-coding RNAs that bind target mRNAs and repress translation, coordinating stress-related pathways. Among them, miR-210 is a well-known hypoxia-responsive marker, whereas miR-181a and miR-206 are associated with myogenesis and growth regulation through MAPK and mTOR signaling. Given the ecological and economic importance and physiological plasticity of Colossoma macropomum, we investigated whether miRNA expression is reversible after exposure to an extreme climate-change scenario. In brain tissue, miR-206 increased 1.2-fold after 30 days of exposure and further to 1.7-fold during recovery. miR-181a decreased 2.4-fold under extreme conditions and remained downregulated (1.9-fold) after recovery. miR-210 was unchanged during exposure but increased 2.8-fold during recovery. These findings indicate partial, miRNA-specific reversibility, suggesting dynamic epigenetic regulation during and after climate stress.

SENSING Sensing the environment: Altered waters and changing behaviours

Sensing the environment: Altered waters and changing behaviours

Tyler Buchinger, Andy Dittman and Keith Tierney

Abstract: We aim to bring together junior to established researchers in the field of fish sensory biology to link sensory signals to fish receivers in order to understand the future of fish populations in the context of ecology and a changing environment. This will be the seventh offering of this symposium. Past symposia have focused on vision, olfaction, taste, hearing, lateral line, magnetic and electroreception and the role they play in the physiology, behavior, and ecology of fishes. We encourage fish sensory biologists from all disciplines (physiology, ecology, behavior) and all senses to participate in the symposium and especially researchers in early career stages and that represent the diversity of people studying how fish investigate the world. For the 2026 Symposium, we especially encourage presentations that focus on the role of sensory systems in the context of the changing coastal environment that fish experience and the use of multiple sensory modalities to adapt to these changes.

SENSING Sensing the environment: Altered waters and changing behaviours

Tuning out the noise: the role of habituation to underwater sound in freshwater fishes

Beneteau, Gabrielle (*beneteag@uwindsor.ca*) Oral Presentation
Co-Authors: Dycha, Grace; Higgs, Dennis
University of Windsor

Abstract: The increase in human-mediated activity has raised concerns regarding the potentially negative impacts of noise pollution on fish populations. Anthropogenic noise impacts the acoustic scene, disrupting fish communication and behaviour, however research is limited in the amount of time that this disruption lasts and whether fish are able to habituate to prolonged exposures. To examine the role of noise habituation, the current study examines behavioural and morphological metrics of noise impacts in free-swimming freshwater fish living in areas that differ in anthropogenic noise profiles. Behavioural experiments were conducted for natural swimming behaviours and in response to sound playbacks, while also quantifying hair cell density in multiple species at each site. Preliminary results of behavioural experiments demonstrate location-specific differences on startle behaviour in natural settings and in response to sound playback. In addition, there were location-dependent differences in hair cell density that correlated with the observed behavioural differences. For all three species, there were significant differences in hair cell density based on epithelial location, with edge locations having higher densities than those in the centre of the epithelium. While results are still preliminary, it appears that sound level in a particular habitat may affect both natural and induced behaviours, suggesting long-term effects of sound.

SENSING Sensing the environment: Altered waters and changing behaviours

Polystyrene nanoplastics damage lateral-line neuromasts and impair flow-guided behaviors across zebrafish life stages

Gong, Chenxin (*22417005@zju.edu.cn*) Oral Presentation
Co-Author: Shi, Wei
Zhejiang University

Abstract: Although plastic-derived nanoplastics are emerging contaminants in aquatic ecosystems, their impacts on mechanosensory systems that mediate flow-dependent behaviors remain poorly understood. Therefore, the effects of polystyrene nanoplastics (PS-NPs) on lateral-line neuromasts and flow-guided behaviors were investigated across zebrafish life stages in this study. Adult zebrafish were exposed to PS-NPs (0.1 and 1 mg/L) for 4 weeks and assessed for rheotaxis, whereas larvae were exposed for 10 days and tested for locomotor stability under vortex-flow conditions. PS-NPs exposure significantly reduced flow-guided performance in both adults and larvae, indicating impaired hydrodynamic sensing. Ultrastructural analyses demonstrated structural degeneration of lateral-line neuromasts, characterized by hair-bundle disruption, kinocilia shortening, increased apoptosis, and reduced hair-cell staining. To elucidate underlying mechanisms, bulk RNA-seq and single-cell RNA-seq of lateral-line cells were also conducted. Transcriptomic analyses indicated mitochondrial and redox dysfunction, disturbed ion homeostasis, and activation of AMPK-mTOR/autophagy pathways following PS-NPs exposure, particularly in sensory hair cells. Immunoblotting further supported the involvement of autophagy- and ferroptosis-associated signaling. In conclusion, our results demonstrate that PS-NPs can impair fish environmental sensing by inducing structural and molecular damage to lateral-line neuromasts, leading to deficits in flow-guided behavior.

SENSING Sensing the environment: Altered waters and changing behaviours

Does hypoxia impair vision in coral reef fish?

Morgan, Rachael[1] (*rachael.morgan@uib.no*) Oral Presentation
Co-Authors: Andreassen, Anna[2]; Campos-Candela, Andrea[1]; Lauridsen, Henrik[3]; Mills, Suzanne[4] and Damsgaard, Christian[3]
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Abstract: Coral-dwelling fish experience large daily fluctuations in oxygen, with hyperoxia occurring during the day and hypoxia building up at night. Nighttime hypoxia can be particularly severe between coral branches where the damselfish Dascyllus aruanus seeks shelter. The retina of the eye has high oxygen demands, so we ask whether the vision of Dascyllus aruanus is impaired by hypoxia, or whether they have evolved mechanisms to retain their vision, ensuring visual detection and reduced predation risk when emerging from the coral. We compare these results to the nocturnal cardinalfish Ostorhinchus angustatus, which we predict is active in more oxygen-rich water, and therefore lacks hypoxia-related adaptations.

Using the optomotor method, we demonstrate that visual acuity declined with progressively severe hypoxia in Dascyllus aruanus but was maintained in Ostorhinchus angustatus. To determine whether this visual impairment arises from insufficient oxygen supply to the eye, we measured retinal oxygen levels in vivo under both fully oxygenated and hypoxic conditions. Retinal oxygen was reduced in Dascyllus aruanus under hypoxia, consistent with the optomotor results. Surprisingly, retinal oxygen levels were also reduced in Ostorhinchus angustatus despite its vision remaining unaffected. To explain this discrepancy, we quantified anatomical properties of the eye and found that damselfish have a thicker retina with greater oxygen demands co-occurring with a larger choroid rete. These adaptations may leave Dascyllus aruanus more vulnerable when environmental oxygen falls. Together, our results point us towards the existence of a significant trade-off between high visual capabilities and the risk of predation during hypoxic events.

SENSING Sensing the environment: Altered waters and changing behaviours

Cholinergic mucocytes evaluated as candidates for O2 sensors in rainbow trout gills

Brink, Deidre (*freerangedee@gmail.com*) Oral Presentation
Co-Authors: Milsom, William
The University of British Columbia

Abstract: O2 sensing and gas exchange occur in fish gills. In rainbow trout (Oncorynchus mykiss) gills, sensory nerve activity from gill arch 1 responded strongly to hypoxic perfusate or ACh (Burleson and Milsom 1995), implicating involvement of at-the-time unidentified cholinergic cells in acute hypoxia sensing. Consequently we mapped cholinergic cells and nicotinic ACh receptors (nAChRs) in unsectioned trout gills using high resolution 3-D fluorescence light microscopy and immunohistochemistry (IHC). Epithelial mucocytes in the trout gill were positive for multiple markers of cholinergic cells: the vesicular ACh transporter, synaptic vesicles, choline acetyltransferase, and nicotinic ACh receptors. Mucocytes were innervated by fine dendrites which extended from cholinergic neurons located at the base of each filament. Most of these features are present in mammalian airway mucocytes. Further, we assessed activity levels in living gill mucocytes by measuring vesicular uptake of the fluorescent styrl FM dyes and AI-based cell segmentation and fluorescence quantification. Relative to aerated perfusate, mucocytes had significantly more dye uptake in acute response to depolarizing perfusate containing high external [KCl], as well as hypoxic perfusate. Assessment for effects of nicotine on FM labeling are currently being measured, as well as the interactive effects of the neonicotinoid nAChR receptor modulator Imidacloprid and acute hypoxia. Recent evidence established that cholinergic neuronal cells in the zebrafish gill (A-NECs) contribute to O2 sensing (Reed et al 2025). Mucocytes may similarly function as O2 sensory cells. Altogether, these data are relevant to both O2 sensing and non-neuronal cholinergic signalling (NNCS) by the innate immune system in teleosts.

SENSING Sensing the environment: Altered waters and changing behaviours

Do group dynamics shape how fish sense and respond to warming temperatures?

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Abstract: Social context is a crucial yet understudied component of an animal's environment. Fish exhibit a variety of social behaviours that could influence their response to an environmental challenge like temperature increases. Adding environmental complexity can further complicate social behaviours. Mangrove rivulus (Kryptolebias marmoratus) is an amphibious fish that exist in diffuse groups and exhibit naturally aggressive behaviours. We assessed whether group size and environmental complexity (e.g., shelter access) affect how fish respond to acute warming using an isogenic lab population of mangrove rivulus. We capitalized on the fish's propensity to voluntarily emerge from water by measuring their voluntary emersion temperature. Individual fish emerged at significantly warmer temperatures (39.4 ± 0.17 °C) than fish in groups of three (38.7 ± 0.07 °C) and compared to individual fish with shelter (38.9 ± 0.12 °C). To assess whether this higher thermal avoidance threshold in individual fish is linked to changes in thermal perception, we titrated capsaicin – the active compound in chili peppers - into the behavioural arena to agonistically target thermosensing transient receptor potential vanilloid 1 (TRPV1) channels and induce emersion with chemical heat. Preliminary results suggest emersion from water occurs at higher capsaicin concentrations when fish are exposed individually without shelter relative to all other treatments. These results are analogous to our heating data and suggest that solitary fish without environmental complexity are least sensitive to warming, likely through a desensitization of their TRPV1 channels. This highlights the importance of group dynamics in shaping how fish will respond to environmental challenge.

SENSING Sensing the environment: Altered waters and changing behaviours

Transient loss of olfaction in brown trout (Salmo trutta) following exposure to dissolved organic carbon: Behavioural, physiological, and cellular responses

Zink, Lauren[1] (*lauren.zink@zoology.ubc.ca*) Oral Presentation
Co-Authors: Tilley, Molly[2]; Ruhr, Ilan[3]; Bogard, Matthew J.[2]; Wiseman, Steve[2] and Wood, Chris[1]
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Abstract: Fish rely on olfactory perception to mediate a range of crucial life-history behaviours. While some work has been done assessing the effect of commercially available humic acid (a component of dissolved organic carbon [DOC]) on olfaction, natural DOC has not been thoroughly assessed. The concentrations of DOC are increasing globally, driven by land use changes, climatic shifts, and recovery from acid deposition; therefore, it is important to assess how DOC concentrations alter fish physiology. By exposing brown trout to 10 mg/L of natural DOC derived from either maple leaves or

aquatic snail cultures, we demonstrate that the gill and olfactory epithelia respond independently, and differently to DOC. We assessed the olfactory acuity of fish using electro-olfactography (EOG), which entailed exposing the fish to DOC, while simultaneously presenting them with a mixture of two odourants, L-alanine and TCA, that stimulate olfactory sensory neurons and produce a measurable electrical potential. Despite the presence of the odourants, we observed an immediate loss of olfactory acuity with the pulse exposure of DOC, but olfaction recovered under sustained DOC exposure. This response was consistent when either odourant was tested independently. We correlated this physiological finding using an open-field behavioural assay and found that predator response behaviour (using TCA as a cue) mirrored the trends observed in EOG. A histological analysis revealed that a mucosal response occurs at the olfactory epithelium when exposed to DOC in which goblet cells empty their contents, providing a potential mechanism for the rapid recovery of olfactory acuity (NSERC, JEB Fellowship).

SENSING Sensing the environment: Altered waters and changing behaviours

Escaping from danger: Neural mechanisms underlying olfactory aversive behaviours in zebrafish

Yoshihara, Yoshihiro (*yoshihiro.yoshihara@riken.jp*) Oral Presentation
RIKEN Center for Brain Science

Abstract: Many olfactory cues pervade the aquatic environment of fish and evoke various olfactory behaviours important for their survival and species preservation, such as searching for foods, escaping from danger, finding potential mates, and memorizing physiologically important odor-associated contexts. Zebrafish has become one of the most popular and useful model organisms in neurobiology. In addition to its general advantageous properties (external fertilization, large clutch size, rapid development, transparency of embryos, etc.), zebrafish is amenable to various genetic engineering technologies including transgenesis, mutagenesis, gene editing, and transposon-mediated gene transfer. The zebrafish genome harbors ~300 olfactory receptor genes consisting of ~140 ORs, 6 V1Rs, ~50 V2Rs, and ~100 Taars. “One neuron-one receptor rule” and “glomerular convergence of like axons” are essentially preserved in zebrafish, resulting in formation of “odor map” on the glomerular array of the olfactory bulb, based on chemical structures of odorants and pheromones. The mitral cells in the olfactory bulb project axons to four major target areas in the forebrain in distinct patterns, so that the odor information is processed differentially in these higher olfactory centers. Thus, the overall circuit organization and the information processing logic in fish are remarkably similar to those in mammals. In this presentation, I will summarize recent advances in our knowledge on the functional architecture of the neural circuits for odor-evoked behaviours in zebrafish. In particular, I will focus on molecular, cellular and circuit mechanisms underlying olfactory aversive behaviours from dangerous chemicals in environment, such as alarm pheromones, carbon dioxide and ammonia.

SENSING Sensing the environment: Altered waters and changing behaviours

Pheromone behavioral antagonists reduce invasive sea lamprey spawning activity in a natural stream during 24-hour trials

Scott, Anne (*scottan7@msu.edu*) Oral Presentation
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Michigan State University

Abstract: Sea lamprey (Petromyzon marinus) are an invasive parasite in the Laurentian Great Lakes that contributed to the historic collapse of the native fish communities. Ongoing control efforts including lampricides and migration barriers are necessary to control sea lamprey. Inherent limitations of these effective strategies highlight the need for supplemental strategies. Sea lamprey rely heavily on chemical cues to locate spawning streams, find mates, and reproduce, which provides an opportunity to disrupt reproduction by manipulating pheromone mediated behaviors. Previous work identified petromyzonol sulfate (PZS) and petromyzonol tetrasulfate (3sPZS) as pheromone behavioral antagonists that reduced olfactory responses and female attraction to the male sex pheromone 3-keto petromyzonol sulfate (3kPZS) over short exposure periods. Here, we evaluated whether a mixture of PZS and 3sPZS reduces spawning activity over a longer timescale in a natural spawning ground. During 24-hour trials conducted across the spawning season, application of the antagonist mixture reduced the total number of spawning events by 53% compared to control trials. Wild female abundance in the system was also lower during antagonist treatments based on visual surveys. Passive integrated transponder (PIT) data from mature male and female sea lamprey released into the spawning stream revealed individuals spent an average of 26% less time in the prime spawning zone of the river when antagonists were applied than during control trials. These results demonstrate that behavioral antagonists can suppress spawning activity over ecologically relevant timescales and provide insights that may inform the development of additional tactics to supplement current control of invasive sea lamprey.

SENSING Sensing the environment: Altered waters and changing behaviours

Neural and sensory bases of social behavior in African cichlids: from mate choice to parental care

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Abstract: Pheromonal sensation is central to social communication, yet their neural and behavioral functions remain incompletely understood in many vertebrates. The African cichlid Astatotilapia burtoni provides a powerful genetic and behavioral model to dissect how olfaction shapes social behaviors across mating and parental care contexts. In the reproductive domain, females release PGF2α-derived compounds that signal fertility to males. Using dichotomous choice assays, we show that males preferentially respond to reproductive female odors, demonstrating a critical role for olfaction. CRISPR-mediated

disruption of olfactory sensory neuron (OSN) subtypes reveals that ciliated OSNs, but not microvillus OSNs, are required for detecting these cues. Specifically, mutation of the ciliated OSN transduction channel cnga2b impairs courtship and pheromone sensitivity, whereas trpc2b mutants do not. We further identify a subset of ciliated OSNs expressing the β-olfactory receptor Or113a that respond to reproductive odors. Unexpectedly, cnga2b and or113a mutant males retrieve eggs during spawning, an unprecedented behavior in this mouthbrooding species, indicating that ciliated OSN signaling mediates mating preference and suppresses male parental behavior. Extending this framework to parental care, we examine how olfaction contributes to parent-offspring recognition during maternal mouthbrooding. Behavioral assays reveal that fry retrieval becomes increasingly selective over the brood period, with late-stage females better distinguishing their own from foreign fry. Using Tol2-mediated transgenesis and optogenetic silencing of defined OSN subtypes, we aim to test the causal roles of these circuits in parental-offspring recognition. Together, our work establishes A. burtoni as a tractable vertebrate model linking olfactory sensation to the neural control of mating and parental care.

SENSING Sensing the environment: Altered waters and changing behaviours

Pheromonal communication mediating courtship behaviour in zebrafish (Danio rerio) is impaired by environmentally relevant total ammonia nitrogen (TAN) concentrations

Rahman, Emir[1] (*erahman@uoguelph.ca*) Oral Presentation
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Abstract: Conspecific sex pheromones act as powerful olfactory signals that initiate and coordinate spawning interactions in many teleost fish. However, increasing nitrogen pollution in aquatic environments raises concerns about how poor water quality may interfere with sensory-mediated reproduction. We previously observed that environmentally relevant concentrations of total ammonia nitrogen (TAN; 2 mg/L) decrease fertilized egg production and suppress key courtship behaviours (chasing, touching, encircling) in zebrafish. We hypothesized that TAN impairs reproductive success by disrupting the olfactory detection and processing of sex-specific pheromones. To test this, we measured behavioural responses (position, orientation, and movement) of male and female zebrafish exposed to 2 mg/L TAN or control conditions following the release of sex-specific pheromones: Prostaglandin $F_2\alpha$ ($PGF_2\alpha$) and 15-keto $PGF_2\alpha$ in males, and 4-androstenedione in females. Control fish of both sexes showed attraction to their respective pheromones, spending more time in the third of the tank near the odour source, and males increased their orientation toward the stimulus. A 12-hour exposure to 2 mg/L TAN significantly reduced male responses to prostaglandin cues, while females showed no attraction to 4-androstenedione. TAN-exposed males also displayed higher locomotor activity regardless of pheromone presence. These findings demonstrate that nitrogen pollution can disrupt pheromone-mediated olfactory detection and alter courtship behaviour in zebrafish. By impairing sensory communication, altered water chemistry may

inadvertently decrease reproductive success, with important implications for fish populations in changing aquatic environments.

SENSING Sensing the environment: Altered waters and changing behaviours

Mechanisms of Neuropeptide Y (NPY) mediated modulation of olfactory sensory neurons in fed and fasted lake sturgeon (Acipenser fulvescens)

Edwards, Tyler[1] (*edward32@myumanitoba.ca*) Oral Presentation
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Abstract: Olfaction plays a critical role in how animals perceive their environment and is important for feeding behaviour. Although the sensory mechanism underlying food detection in vertebrates are well characterized, the interaction between the olfactory system and endocrine pathways that regulate food intake is not fully understood. In both mammals and fishes,food intake is typically controlled by satiety hormones produced by central regions such as the hypothalamus, as well as peripheral tissues including the gastrointestinal tract. The objective of this study was to investigate the mechanism underlying NPY mediated modulation of OSNs in lake sturgeon. Patch-clamp electrophysiology was used to characterize the biophysical properties of OSNs, while in situ hybridization chain reaction (ISHCR) was used to localize expression of NPY and NPY Y1 receptor in the olfactory epithelium of fed (ad-libitum) andfasted (14 days) fish. Biophysical properties, including first action potential thresholds, latency, slope and spike analysis, were quantified in the presence and absence of NPY (n = 15 per dietary state per treatment). ISHCR was performed using probes for OSN subtype markers (Microvillous; TRPC2 & V2R1, Ciliated OMP & CNGA2) as well as NPY and the NPY Y1 receptor, to provide cellular context and determine receptor localization (n = 4 per dietary state). This work will elucidate mechanisms underlying hormonal modulation of olfactory function and establish a framework linking sensory physiology to dietary and metabolic state at single-cell resolution.

SENSING Sensing the environment: Altered waters and changing behaviours

Impacts of early rearing substrate on the olfactory system development and function in chum salmon (Oncorhynchus keta)

Sevova, Ralitza[1] (*ralitza.sevova@zoology.ubc.ca*) Oral Presentation
Co-Authors: Bouyoucos, Ian[1]; Reich, Eric[1]; Leitch, Duncan[2]; Brauner, Colin J.[1] and Matthews, Ben[1]
[1]University of British Columbia; [2]University of California Los Angeles

Abstract: In aquatic environments, olfaction underpins essential behaviours including predator avoidance and detecting food sources. In chum salmon (Oncorhynchus keta), olfactory imprinting during early development is important for successful homing, which is critical for population stability and genetic diversity. Despite conservation efforts,

hatchery-reared salmon are typically raised in barren tanks that differ markedly from natural riverbeds. While economically efficient, these environments can negatively impact sensory and neural development. Hatchery-reared fish often exhibit smaller olfactory bulbs, increased stress and aggressive behaviours, and a reduced brain and body size compared to their wild conspecifics. We hypothesized that early-life substrate enrichment will increase olfactory neural activation and behavioural responsiveness in chum salmon. To test this, fish were reared to the fry stage under four conditions: no substrate (control), river rock, gravel, and BioBalls. Neuronal activation within the telencephalon was quantified after exposure to taurocholic acid, a potent bile acid odourant, using immunohistochemistry with c-Fos as a marker of immediate early gene activation. Behavioural sensitivity was assessed through measuring avoidance responses to a whole-body extract that acted as an alarm cue. We predicted that substrate-enriched fish would exhibit increased c-Fos expression in olfactory regions and stronger behavioural responses compared to fish reared in barren tanks. By linking early environmental structure to neural activation and olfactory behaviour, this work aims to inform hatchery practices and improve the ecological relevance of rearing environments for conservation-focused fisheries management.

SENSING Sensing the environment: Altered waters and changing behaviours

Ocean acidification disrupts olfactory signal transduction and receptor structure in the large yellow croaker

Shi, Wei (*shiwei1992@zju.edu.cn*) Oral Presentation
Co-Authors: Gong, Chenxin
Zhejiang University

Abstract: Ocean acidification (OA), driven by rising anthropogenic CO_2 emissions, has caused an unprecedented decline in seawater pH, threatening marine ecosystems and fisheries. Olfaction is a critical sensory system in fish, mediating essential behaviors such as foraging, predator avoidance, migration, and reproduction. Because the olfactory epithelium (OE) is directly exposed to seawater, it is particularly vulnerable to environmental stressors. However, the mechanisms underlying OA-induced olfactory dysfunction remain poorly understood.In this study, we investigated the effects of OA on the olfactory system of the large yellow croaker (Larimichthys crocea), an economically important mariculture species in China. Electro-olfactogram recordings showed that OA exposure significantly reduced olfactory responses. Histological and molecular analyses revealed structural damage to the OE, increased apoptosis, and decreased expression of olfactory marker protein (OMP), G protein-coupled receptors (GPCRs), and ion transport activities, indicating impaired odorant detection and signal transduction. Furthermore, cell-free expression of the croaker olfactory receptor protein followed by 1H NMR and circular dichroism analyses demonstrated pH-dependent conformational alterations, suggesting that reduced pH directly compromises receptor structure and ligand-binding capacity. Comparative transcriptomic analyses further revealed significant alterations in pathways related to apoptosis, signal transduction,energy metabolism, and neural

function.Overall, our findings demonstrate that OA impairs olfactory perception of L. crocea by inducing OE damage and apoptosis, suppressing receptor function, and directly altering receptor conformation under acidic conditions.

SENSING Sensing the environment: Altered waters and changing behaviours

Evidence of wildfire-driven differential gene expression in juvenile steelhead olfactory epithelium

Abbott, Morgan M[1]. *mmabbott@ucsc.edu* Oral Presentation
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Abstract: Wildfires can mobilize heavy metals and other contaminants into streams, yet the effects of wildfire disturbance on the sensory biology of aquatic organisms remain poorly understood. Juvenile steelhead (Oncorhynchus mykiss) rely on olfaction for predator avoidance, foraging, and natal stream imprinting, and disruption of olfactory function could reduce individual fitness and population persistence. The 2020 wildfires along California's Central Coast provided a paired watershed natural experiment comparing burned and unburned reaches across four watersheds. We used RNA-sequencing of olfactory epithelial tissue from 80 juvenile steelhead across four watersheds (10 burned, 10 unburned per watershed) to test whether wildfire disrupts the expression of important olfactory genes. We identified 2,179 differentially expressed genes globally, with 202 to 5,963 significant genes within individual watersheds. Comparative genomics against zebrafish and Atlantic salmon proteomes revealed that differentially expressed genes included an olfactory receptor gene upregulated in burned fish across three of four watersheds, multiple interferon and immune signaling genes consistently downregulated in burned fish, a metal transporter relevant to heavy metal exposure, and stress response and detoxification genes. The immune suppression signal was the dominant transcriptomic response. The identification of a differentially expressed olfactory receptor provides novel field-based evidence of wildfire-driven disruption of olfactory gene expression in wild salmonid populations. These findings have implications for understanding how wildfire affects chemosensory function critical to predator avoidance, natal homing, and population recovery in fire-affected landscapes.

SENSING Sensing the environment: Altered waters and changing behaviours

Behavioral Responses of Cisco (Coregonus artedi) to Putative Imprinted Odors and Conspecific Odors

Gill, Hannah[1] (*gillhann@msu.edu*) Oral Presentation
Co-Authors: Honsey, Andrew[2]; Johnson, Nicholas[2]; Gordon, Roger[3] and Buchinger, Tyler[1]
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Abstract: Many fish are thought to use imprinted odors and conspecific odors to navigate to spawning sites and find mates. Pacific salmon are a well-studied example that imprints to the odor of their natal site during early life stages and later uses these odors to home to spawning sites, with odors from other conspecifics potentially acting as secondary cues. Though olfactory imprinting has been proposed as a mechanism for natal homing in many fish species, few studies have investigated imprinting outside of Pacific salmon and differences in life histories among species raise uncertainty about generalizing the conceptual model of olfactory imprinting based on Pacific salmon. Here, we investigated the responses of cisco to odor cues suspected to guide fish to spawning grounds and potential mates. We hypothesize cisco use imprinted odors as primary cues and conspecific odors as secondary cues to locate spawning grounds and conspecifics. In two-current choice flume behavioral assays, hatchery-raised male cisco preferred the water they were reared in up to 16 months post-fertilization (putative imprinted odor) over a negative control during the two weeks before spawning and male conspecific odor over a negative control during active spawning. These results support our hypothesis that cisco, a non-Pacific salmonid, use putative imprinted odor and male conspecific odor cues during spawning, but suggest cisco prioritize different odor cues during different phases of spawning. Future work is needed to understand the role of these cues in context to cisco life history. These findings will help inform the restoration of cisco in the Laurentian Great Lakes.

SENSING Sensing the environment: Altered waters and changing behaviours

Transcriptional responses of the olfactory system of Pacific salmon to aquatic acidification

Bouyoucos, Ian[1] (*ian.bouyoucos@zoology.ubc.ca*) Oral Presentation
Co-Authors: Reich, Eric[1]; Sevova, Ralitza[1]; Leitch, Duncan[2]; Matthews, Ben[1] and Brauner, Colin J.[1]
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Abstract: Olfactory imprinting, the development of chemical memories that guide spawning migrations to natal habitats years later, is a hallmark trait of Pacific salmon (Oncorhynchus spp.). Not all migrating adults return to their natal stream to spawn, and one potential underlying cause is alterations to the water chemistry larval and juvenile salmon experience while imprinting. Carbon dioxide (CO2) is known to impair olfaction in fishes; however, whether it affects olfactory imprinting is not yet known. Here, we tested the hypothesis that CO2 can impair olfactory imprinting and homing through effects on the peripheral and central nervous system during two putative developmental windows for imprinting, yolk-sac absorption (YSA) and the parr-smolt-transformation (PST). We predicted that exposing Pacific salmon to elevated CO2 during YSA and PST would downregulate mRNA transcripts of important receptors, olfactory genes, and neural markers in the olfactory epithelium, olfactory bulb, and telencephalon. Chinook (O. tshawytscha), coho (O. kisutch), and pink (O. gorbuscha) salmon were obtained as unfertilized gametes or as parr and were exposed to CO2 tensions representative of natural

aquatic acidification or groundwater used in salmon hatcheries. Chinook were exposed to CO2 (2,000 µatm) from fertilization through PST or only PST, coho were exposed to CO2 (1,000 or 2,000 µatm) during PST, and pink salmon were exposed to CO2 (2,000 or 5,000 µatm) and an imprinting cue (phenylethyl alcohol) as eyed eggs until YSA. Together, these experiments provide a comprehensive look at the potential for aquatic acidification by CO2 to impair critical olfactory-mediated behaviours in Pacific salmon.

SENSING Sensing the environment: Altered waters and changing behaviours

Impact of CO_2 on Pacific salmon olfaction during key imprinting periods

Reich, Eric[1] (*eric.reich@zoology.ubc.ca*) Oral Presentation
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Abstract: Pacific salmon (Oncorhynchus spp.) exhibit an incredible homing ability that is achieved through olfactory imprinting during freshwater juvenile life stages. Due to a decline in salmon populations, hatcheries have been implemented to supplement wild stocks; however, hatchery raised fish exhibit increased straying behaviors, more often migrating to alternative spawning sites. One potential cause of this behavior is an impairment in imprinting mechanisms when juveniles begin their ocean migration (smoltification). This impairment may be due to higher-than-normal CO_2 tensions in hatchery conditions, ranging from 400-5000 µatm. Previous studies indicate a CO_2 tension of 2000 µatm may impact olfactory receptor sensitivity to amino acid agonists in Pacific salmon. We hypothesize that straying behaviors may be exacerbated by high CO_2 tensions in hatcheries altering olfactory receptor sensitivity and thus memory and homing behaviors. To examine this, we exposed pink salmon alevin (O. gorbuscha) and coho parr (O. kisutch) to 2000 µatm CO_2 during which we introduced odourants, a combination of phenylethyl alcohol (PEA) and L-arginine, over imprinting periods. We assessed mRNA expression of olfactory receptors (ORs) or300-1 and basic amino acid receptor (baar) in the olfactory epithelium. We found a decrease in OR expression in the presence of the odourant combination while the effect of CO_2 was negligible. Post-smoltification, we subjected coho to choice flume behavioral trials finding no noticeable difference between conditions. These results give insight into the impact of CO2 on Pacific salmon behavior and expression of various ORs during key imprinting periods.

SENSING Sensing the environment: Altered waters and changing behaviours

Fixing an absence of evidence in chemosensory landscaping

Tierney, Keith (*ktierney@ualberta.ca*) Oral Presentation
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Abstract: Freshwater systems contain an incredible diversity of chemicals that vary over space and time. These chemicals arise from myriad natural sources and differ considerably in their solubility and half-life. It is probable that many, if not most, chemicals are detectable by olfactory sensory neurons in the fish nose, owing to the diversity and complexity of olfactory sensory neurons (OSNs). We are interested in determining if chemicals released by organic decay and bacteria may be detectable by OSNs, and if so, could serve as the basis for long-term olfactory memory. Our motivation in this was the role of odorant bouquets in guiding salmonid migration, which largely remains enigmatic, in part because recent studies cast doubt on the role of single classes of compounds. We will present information on the ability of untested but conceivably relevant dissolved chemicals to serve as odorants, using the zebrafish model. We aim to demonstrate that inter-kingdom molecules may function as local olfactory landmarks that facilitate wayfinding in migratory fishes.

SENSING Sensing the environment: Altered waters and changing behaviours

Changes in odorant receptor gene expression in naturally migrating adult Chinook salmon exposed to amino acid odorants as embryos.

Dittman, Andrew[1] (*andy.dittman@noaa.gov*) Oral Presentation
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Abstract: Salmonids are renowned for their reproductive homing migrations from oceanic feeding grounds back to their freshwater natal sites. This remarkable navigational ability is governed, at least in part, by olfactory discrimination of home-stream odors learned (imprinted upon) prior to and during their seaward migration as juveniles. Controlled experiments have demonstrated specific changes in olfactory receptor mRNA expression coincident with imprinting to amino acid odorants.Furthermore, these gene expression changes have been linked to discrete, sensitive developmental periods for imprinting, as well as during the period of maturation and homing in laboratory studies. Imprinting and homing in a natural environment, with constantly varyingstimuli from changing environmental conditions and habitats, is likely much more complex. To examine whether naturally migrating salmon would exhibit similar olfactory receptor expression changes as laboratory subjects, we exposed hatchery Fall Chinook salmon to an amino acid odor cocktail or control hatchery water at distinct developmental stages and examined changes in odorant receptor gene expression in olfactory rosettes prior to release from the hatchery

and upon return as adults. During the adult return migration, imprinting amino acids were metered into the adult return ladder in an effort to increase homing fidelity.Elevated odorant receptor mRNA expression was observed in returning adults that had been exposed to amino acid odorants as embryos prior to release, relative to odorant-naive fish. We discuss these findings in the context of odor-exposure timing and homing fidelity of experimental fish to their natal hatchery.

SENSING Sensing the environment: Altered waters and changing behaviours

Hypoxia and CO2/pH responses of S-NECs and gill raker chemosensory cells in rainbow trout (O. mykiss) gills

Brink, Deidre (*freerangedee@gmail.com*) Poster Presentation
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Abstract: S-NECs (serotoninergic neuroepidermal or neuroendocrine cells) are found on gills of most species of fish and are viewed as the primary sensory cells for hypoxia and possibly CO2/pH sensing. They are believed to be epidermal and lie in close or immediate contact with environmental water. Using 3-D light microscopy we show that S- NECs have complex shapes and are sub-epidermal to mucocytes in fixed filaments labelled with an anti- serotonin antibody and the mucin/mucocyte marker soybean hemagglutinin. Similarly we found that living FM 4-64 labelled mucocytes ex-vivo in rainbow trout gills were external to sub-epidermal S-NECs labelled with the serotonin transporter permeable dye APP+. S-NECs were positive for nicotinic ACh receptors suggesting sensitivity to ACh. S-NECs concentrated APP+ at similar levels in aerated perfusate, weakly depolarizing 2[KCl]ex ,and hypoxia. However, perfusate containing CO2 significantly enhanced APP+ fluorescence in NECs relative to other conditions, suggesting that the S-NECs were more responsive to CO2/pH than hypoxia. Using similar 3-D imaging protocols, we also found clustered fusiform cells similar to the chemoreceptive taste cells found in zebrafish and other species within the gill rakers. These cells were positive for ZN-12/HNK1, were innervated, but were immuno- negative for serotonin, vesicular acetylcholine transporter, and tyrosine hydroxylase. They rapidly and reversibly labelled with statistically similar intensities in aerated perfusate or weak depolarization in 2[KCl]ex , but labelled significantly more intensely in 1 mm NaCN or hypoxia. These data support the perspective that there are multiple sensory pathways contributing to respiratory and environmental gas sensing.

SENSING Sensing the environment: Altered waters and changing behaviours

Integrating automated behavioural tracking into standardized fish avoidance assays to expand behavioural endpoints

Castro Barquero, Pablo (*pablocb2578@gmail.com*) Poster Presentation
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Abstract: Behavioural endpoints are increasingly recognized as sensitive indicators of sublethal contaminant exposure in aquatic organisms. In ecotoxicology, standardized avoidance assays such as the Heterogeneous Multi-Habitat Assay System (HeMHAS) are commonly used to evaluate fish responses to environmental contaminants by measuring habitat selection and avoidance behaviour. These assays provide valuable information on the ability of organisms to detect and avoid polluted environments; however, most analyses focus primarily on spatial distribution outcomes while other behavioural responses occurring during the experiment remain largely unexplored.

Recent advances in automated video-tracking software provide new opportunities to expand the analytical potential of these experiments. Tools such as AnimalTA allow detailed extraction of movement and activity data from video recordings, including locomotor activity, travelled distance, spatial occupancy, zone transitions and exploration patterns. These analyses are particularly suitable for experiments involving individual organisms or small groups, where accurate tracking of movement can be achieved.

Here we propose the integration of automated behavioural tracking as a complementary analytical approach within standardized avoidance assays. By applying automated tracking to complementary trials conducted under the same experimental arenas and environmental conditions used in HeMHAS experiments evaluating fish responses to copper exposure, it becomes possible to obtain multiple behavioural endpoints alongside traditional avoidance metrics.

This approach represents a low-cost and time-efficient strategy to increase the amount of behavioural information generated from ecotoxicological experiments. The development of standardized behavioural datasets may contribute to establishing reference behavioural responses that complement traditional avoidance metrics, ultimately improving the sensitivity and ecological relevance of behavioural ecotoxicology.

SENSING Sensing the environment: Altered waters and changing behaviours

Interregional comparison of gobiid fauna associated with alpheid shrimp burrows

Tamaki, Shotaro[1] *tamasho-0605@docomo.ne.jp* Poster Presentation
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Abstract: Japan spans a wide latitudinal range and is surrounded by ocean, resulting in diverse marine environments. The Pacific coast is heavily influenced by the Kuroshio Current, whereas the Sea of Japan coast exhibits relatively small tidal variations. These contrasting oceanographic conditions support a wide variety of coastal marine organisms,

which in turn lead to diverse interspecific interactions. In Japan, the symbiosis between pistol shrimp Alpheus brevicristatus and gobiid fishes is well known; however, the specific goby species that utilize shrimp burrows and how their species composition varies across regions and environments remain unclear. In this study, we investigated sandy–muddy shores in the Oki Islands (Shimane Prefecture) and a tidal flat in Takehara City (Hiroshima Prefecture) to clarify the species composition of gobies associated with the burrows of A. brevicristatus and to observe their symbiotic behavior. Our results indicated that the primary gobiid species using the burrows at both sites was Acentrogobius virgatulus. However, the composition of symbiotic gobies differed between the two locations, suggesting that shrimp–goby associations may shift in response to local environmental conditions, such as water temperature or surrounding communities. Behavioral observations further revealed that A. virgatulus spent most of its time inside or near the shrimp burrow, indicating a high degree of dependence on the burrow and a strong association with the alpheid shrimp.

SENSING Sensing the environment: Altered waters and changing behaviours

Olfactory Gene Expression During Three Juvenile Life Stages in Fall Chinook

Obley, Miriam[1] (*obleym@oregonstate.edu*) Poster Presentation
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Abstract: Natural-origin salmon imprint on odors in their rearing environment which enables them to navigate back to their natal streams to spawn. Hatchery-reared fish may miss out on specific olfactory cues due to lack of imprinting which may lead it increased straying behavior. The purpose of this study is to examine how olfactory gene expression changes throughout the juvenile life stage in Fall Chinook. Hatcheries typically expose fish to river odors during the parr-smolt transformation, as this period has been shown to be key in olfactory imprinting. However, there is also evidence that juvenile salmon imprint at multiple life periods including as embryos. It is possible that salmon imprint on odors unique to areas in a river at different life stages, creating olfactory waypoints for future navigation. This would indicate that multiple life stages are critical for spawning site fidelity and should be considered when exposing fish to odors in a hatchery. We exposed three groups of Fall Chinook to river water and cyprinid water at three different life stages; Embryonic (Jan 23 – Mar 25), Early smolt (May 23 – Jul 25), Late smolt (Aug 21 – Oct 23). Olfactory rosettes were collected from each group throughout and at the end of each exposure period. RNA was isolated from the tissue and is in preparation for RNA sequencing to explore how olfactory gene expression differs among the three groups. Understanding how gene expression changes throughout rearing can help inform hatchery management practices and when to expose fish to odors to mitigate increased straying.

CARDIO Cardiac responses to changing environmental conditions

Cardiac responses to changing environmental conditions

Todd Gillis, Todd and Erika Eliason

Abstract: The heart is central to fish performance, circulating blood gases, nutrients, hormones, immune factors, and metabolic wastes throughout the body. This symposium highlights research examining how the heart responds and adapts to changing environmental conditions. We invite talks that explore cardiac function across levels of biological organization (gene, cell, tissue, whole animal), and in response to a range of environmental conditions (e.g. temperature, hypoxia, salinity, pH, toxicants). Studies examining responses across different timescales (e.g. acute, chronic, seasonal, fluctuating, generations) are encouraged.

CARDIO Cardiac responses to changing environmental conditions

Myocardial Contraction Kinetics and Performance Underlie Differences in Cardiac Pumping and Metabolic Capacity Between Tropical Fish Species

Gamperl, Anthony[1] (*kgamperl@mun.ca*) Oral Presentation

Co-Authors: Syme, Doug[2]; Seamone, Scott[3]

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Abstract: The scaled sardine, Nassau grouper and schoolmaster snapper are tropical marine species that vary greatly in their thermal tolerance (CTMax = ~ 34.9, 36 and 38.9oC, respectively) and metabolic capacity (~3000, 400 and 1000 mg O2 kg-1 hr-1, respectively). To examine whether differences in temperature- and frequency-dependent effects on heart muscle contractile function can explain these inter-specific differences, we measured the force and power output of ventricular strips from 26oC-acclimated fish at 26, 30 and 34oC (and 38oC in snapper) and at a range of contraction frequencies (30 to >300 min-1). Sardine hearts could be paced at very high frequencies (i.e. heart rates, fH) and had a relative ventricular mass (~0.13%) approximately twice that of the other species. These features, combined with the fact that the fast twitches of the sardine's muscle resulted in a relatively low lengthening power (which allows for adequate diastolic filling at high fH) would clearly support their very high metabolic rate. However, the inherently rapid twitches of their muscle also resulted in extremely brief and low force contractions at warmer temperatures, to the point that their myocardium could not produce power at 34oC. Conversely, the inherently low temperature-dependent and maximum fH of the grouper's myocardium, and the resultant slow twitches which would require a greatly elevated lengthening power to fill the heart, restrict this species' cardiac pumping and metabolic capacity, and thermal tolerance. Finally, the above characteristics are intermediate in the

snapper, and this allows for a moderate pumping (and metabolic) capacity that can be sustained at high temperatures.

CARDIO Cardiac responses to changing environmental conditions

Thermal stress and Lake Sturgeon cardiac cellular function: an electrophysiological study.

Folkerts, Erik [1] (*Erik.Folkerts@umanitoba.ca*) Oral Presentation
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Abstract: Due to the impacts of climate change, temperature is a commonly observed stress experienced in many waterbodies globally. Owing to their ectothermic nature, fish are directly impacted by water temperature changes, and the degree to which fish respond to such changes ultimately decides their fate. The impacts of thermal stress are well documented in many aspects of fish biology, including cardiac performance. However, within the context of species that routinely experience large seasonal variations in habitat temperatures, the impacts of temperature on cardiac function at the cellular level are less known. The impacts of both acute and acclimatory temperature stressors on ventricular cardiomyocyte electrophysiological properties were investigated in the endangered Lake Sturgeon (Acipenser fulvescens). Fish were acclimated to three different, environmentally relevant temperatures (10°C, 15°C, and 20°C). Cellular impacts were assayed via whole-cell patch clamping techniques to record changes in gross electrophysiological function (i.e., action potentials), as well as changes to ion-specific current properties (e.g., current type, density, voltage relationships, etc.). As observed in other fish species, acclimation temperature impacted specific cardiomyocyte electrophysiological properties, including action potential-related metrics and ion current activities (e.g., Ca2+ currents). However, novel combinatorial effects of acute temperature changes were observed in cells across differentially acclimated sturgeon, indicating that certain conditions more impactfully affect this fish's cardiomyocyte function and tolerance to thermal change. Our results have clear physiological and ecological implications given the biology and conservation status of this ancient fish, as well as projected temperature increases across much of the Lake Sturgeon's natural range.

CARDIO Cardiac responses to changing environmental conditions

Embryonic temperature leaves lasting effects in cardiac phenotype in Atlantic salmon (*Salmo salar*)

Johansen, Ida (*ida.johansen@nmbu.no*) Oral Presentation
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Cardiovascular development is highly sensitive to temperature during early development in ectotherms, yet evidence for persistent effects on cardiac phenotype remains limited. Here, we investigated how embryonic and post hatch-temperature influences cardiac structure and function in Atlantic salmon (Atlantic salmon) throughout life. Initial experiments showed that cardiac structure and function are already influenced by egg (2 vs. 8 °C) and post-hatch (10 vs 14°C) temperature in yolk-sac larvae. More specifically, warm reared (8->14°C) larvae had rounder ventricles with higher frequency of cardiac deviations and higher intrinsic heart rate, compared to cold-reared (2->10°C) larvae. Differences in cardiac structure and function persisted throughout freshwater and sea water rearing, indicating that embryonic and later rearing temperature affect cardiac phenotype. To assess whether embryonic temperature alone leaves a lasting impact on cardiac phenotype, two groups with contrasting embryonic rearing-temperature (2 vs. 8 °C) were kept in a common garden throughout the subsequent freshwater and sea water stages. Throughout the freshwater period, few differences in structure and function were observed between these two groups. Pronounced group differences in both cardiac shape and function did however emerge from mid marine phase onwards. More specifically, warm incubated individuals developed progressive ventricular remodelling accompanied by increased contractility and atrioventricular regurgitation at rest. Later, warm incubated fish showed signs consistent with pathological remodelling including myocardial fibrosis and a shift in atrioventricular valve collagen composition consistent with a stiffer phenotype. Together, these findings demonstrate that embryonic temperature has permanent effects on cardiac remodelling and plasticity with long-term consequences for cardiovascular function.

CARDIO Cardiac responses to changing environmental conditions

The role of cardiac remodelling in the acclimation of embryonic and juvenile Sheepshead minnow (Cyprinodon variegatus) to sub-optimal temperatures during embryonic development.

Harman, Adam (*harman@zoology.ubc.ca*) Oral Presentation
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Abstract: The need to assess the thermal tolerance of aquatic species is more apparent as global temperatures continue to rise, however tolerance endpoints are complex and are modulated by a myriad of physiological processes. The relative importance of these physiological mechanisms influencing acute thermal tolerance (i.e. CTmax) are poorly understood. The cardiorespiratory system in fish is morphologically plastic, and remodelling has been shown occur in response to thermal acclimation but has not yet been evaluated in embryos. To investigate the contribution of the cardiorespiratory system to acute thermal tolerance Sheepshead minnow (Cyprinodon variegatus) embryos were incubated at three temperatures (16, 28 and 37°C) until 2 weeks post-hatch. A sub-set of embryos were sampled at Stage 29 for assessment of thermal tolerance using a modified CTmax protocol, and the remaining embryos were kept at the treatment temperature until 2 weeks post-hatch. Samples were collected at hatch for histological analysis of cardiac morphology. CTmax showed plasticity in response to incubation temperature in both embryos and juveniles, with higher incubation temperatures leading to increased thermal tolerance. Physiological data on cardiac remodelling is currently being analyzed.

CARDIO Cardiac responses to changing environmental conditions

The molecular regulation of cardiac remodeling in trout (Oncorhynchus mykiss) caused by thermal acclimation and/or a high salt diet

Robinson, Sean (*swm2robinson@gmail.com*) Oral Presentation
Co-Authors: Gillis, Todd
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Abstract: Rainbow trout (Oncorhynchus mykiss) can undergo cardiac remodeling in response to seasonal temperature. It is hypothesized that cardiac plasticity is triggered by changes in blood viscosity and, consequently, vascular resistance leading to the activation of cell signaling cascades. It has been suggested that these changes are regulated, at least in part by transforming growth factor-β1 (tgfb1) and miR-29b. This study aims to investigate the effect of increased blood pressure on cardiac collagen regulation in rainbow trout and the endogenous regulation of tgfb1 and miR-29b transcript abundance during the initiation of thermal cardiac remodeling. To accomplish this, rainbow trout (n = 84) were fed either a control diet (~1% NaCl: CD) or a high-salt diet (11% NaCl: HSD) for four weeks to induce hypertension. Afterwards, each group were subdivided into three temperature treatments (4 °C, 11 °C, and 18 °C) and fish were acclimated to their target temperatures at a rate of 1 °C per day. Trout were sampled on the third day of temperature ramp-up or ramp-down.

Relative expression of tgfb1 in cold-acclimating trout on the CD was 1.7-fold that in warm acclimating fish and 1.8-fold that in cold-acclimating trout fed the HSD. At 11°C, the high salt diet caused a 1.6-fold increase in the expression of tgfb1. The relative expression of miR-29b in cold-acclimated trout was 2.3-fold and 2.4-fold that compared to that in warm-acclimating trout and cold-acclimating trout fed HSD, respectively. These results support the role of tgfb1 and miR-29b in response to increases in vascular resistance and seasonal temperature remodeling.

CARDIO Cardiac responses to changing environmental conditions

Structural cardiac plasticity in Atlantic bluefin tuna (Thunnus thynnus) under aquaculture and thermal regimes

Wierer, Amelie[1] (*ameliesophia.wierer@imbrsea.eu*) Oral Presentation
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Abstract: Cardiac responses to aquaculture conditions remain understudied in Atlantic bluefin tuna (ABFT, Thunnus thynnus). In other teleosts, such as Atlantic salmon, aquaculture-related stressors can induce morphological deviations and cardiac remodeling. Here, we investigate whether farmed ABFT hearts exhibit morphological and histological differences compared to wild individuals and explore whether these are modulated by temperature.
We analyzed 189 hearts from wild and farmed ABFT (juveniles and adults) collected from 2020-2024. To evaluate thermal impacts, farmed juveniles were reared at two temperature regimes (21 °C and 24 °C). Cardiac morphology was quantified using image analysis (Fiji/ImageJ), focusing on eight shape parameters. Histological sections (HES staining) were used to examine tissue composition, cardiomyocyte nuclei, and structural remodeling. Preliminary results show distinct morphological differences between wild and farmed ABFT. Culture conditions are linked to ventricular remodeling in both adults and juveniles, while additionally altering ventricular symmetry in juveniles. Juveniles cultured at higher temperatures exhibit ventricular remodeling and decreased bulbus arteriosus width. Furthermore, variations in ventricular elongation and bulbus width occur among wild juveniles captured in different geographic areas. Histological analysis is expected to reveal altered tissue characteristics in farmed individuals. Final results will detail quantitative differences. This study presents the first quantitative assessment of the ABFT heart, demonstrating morphological differences between farmed and wild ABFT that already emerge in juvenile stages. Linking cardiac responses to environmental conditions such as temperature may help explain how environmental variability influences cardiac performance in ABFT, providing a physiological basis for assessing resilience and adaptation in the wild.

CARDIO Cardiac responses to changing environmental conditions

Seasonal patterns in the cardiac temperature sensitivity of northern fishes

Williams, Emily[1] (*epwilliams3@alaska.edu*) Oral Presentation
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Abstract: The Arctic is characterized by extreme seasonality. Winter commonly lasts for ~10 months with fish experiencing frigid temperatures (marine: -1.8°C, freshwater: 0-2°C) resulting in distinct physiological challenges. While summer is brief, temperatures can be much warmer (maximum marine: ~13°C and freshwater: ~21°C). Given the extreme nature of this variation and the rapid rate at which the Arctic is warming, Arctic fishes are an intriguing group in which to study how animals cope with changes in environmental temperature. The heart is one vital organ that common changes in response to short and prolonged temperature shifts.Considerable diversity exists among fishes in the response of the heart to temperature variation, but underlying mechanisms and significance are unclear. For example, adrenaline can be critical for maintaining heart function (e.g. heart rate) at temperature extremes, but whether changes in temperature sensitivity are driven by changes in the effect of adrenaline requires exploration. To identify seasonal changes in the thermal sensitivity of heart function and role of adrenaline, we examined the response of heart rate to acute warming in several common fish species at the end of winter and summer in the Canadian Arctic. We found that some species exhibit remarkable flexibility in the temperature sensitivity of the heart (e.g., Greenland cod) while others do not (e.g., lake trout). The Canadian Arctic is among the most rapidly warming regions on Earth. Thus, these differences in the ability to adjust temperature sensitivity may influence the ability of different species to cope with ongoing environmental change.

CARDIO Cardiac responses to changing environmental conditions

Egg size impacts embryonic cardiac thermal performance and growth in Atlantic salmon (Salmo salar)

Morefield, Robert (*robert.morefield@maine.edu*) Oral Presentation
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Abstract: Egg size in Atlantic salmon (Salmo salar) is a maternal trait with a significant amount of adaptive plasticity to environmental conditions related to reproduction. In a hatchery setting, these environmetal influences are absent, providing an excellent opportunity to investigate the role egg size plays on thermal cardiac performance within embryos. We tested how family-level variation in egg size relates to embryonic cardiac thermal performance and early growth in domesticated broodstock lines of Atlantic

salmon reared under standardized hatchery conditions. Using acute warming challenges, we quantified key metrics of thermal performance curves in embryos from multiple families spanning a broad range of egg diameters and tracked larval length at hatch and during early growth. Contrary to prevailing diffusion-based models, embryos from larger-egg families maintained increased heart rates over a wider thermal range, achieved higher breakpoint and peak temperatures, and reached higher maximum heart rates. Egg size was also positively associated with early growth, but not however, with size at hatch. Surprisingly, maternal body mass showed no detectable relationship with any cardiac or growth trait. These results indicate that variation in maternal provisioning, expressed through egg size, can enhance embryonic cardiac capacity and early growth under hatchery conditions, potentially via changes in energetic reserves and egg capsule diffusive properties. Our findings highlight egg diameter as a simple, scalable indicator trait that could be incorporated into selective breeding and conservation programs to help identify broodstock lines producing more thermally robust, high-quality eggs.

CARDIO Cardiac responses to changing environmental conditions

Chronic stress induced cardiac remodeling, depleted energy reserves, and reduced swimming performance of Atlantic Salmon

Opinion, April Grace[1] (*aprilgrace.opinion@uantwerpen.be*) Oral Presentation
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Abstract: Salmonids experience substantial mortality in aquaculture and wild environments. In Atlantic salmon aquaculture alone, mass mortality events have alarmingly increased in frequency and severity, with ~865 million deaths recorded globally between 2012 and 2022. Although the underlying causes remain unclear, mortality peaks often occur following stressful interventions and extreme environmental conditions, with cardiac abnormalities identified as an important risk factor. Our recent meta-analysis revealed deviations in the heart morphology of farmed fish compared to their wild conspecifics. These farming-induced alterations seem maladaptive and have been linked to chronic stress primarily through studies involving exogenous exposure to the stress hormone cortisol. However, a direct experimental link between endogenous cortisol and cardiac pathology has not been established. Here, we exposed Atlantic salmon to unpredictable chronic stress for 30 days and compared their cardiac morphology and performance with undisturbed controls. These cardiac parameters were then correlated with systemic stress indicators. Results showed that stressed fish developed farming-related heart remodelling, including larger, rounder, and less symmetrical hearts, accompanied by reduced cardiac performance as reflected by lower critical swimming speed (Ucrit). Cardiac remodeling appeared linked to sustained increases in workload, indicated by elevated heart rate under chronic stress. While basal glucose and lactate levels were unaffected, stressed fish showed smaller post-Ucrit increases in these metabolites and a reduced hepatosomatic index, suggesting depleted energy reserves that

likely contributed to impaired performance. Unexpectedly, plasma cortisol levels did not differ between treatments, indicating that long-term stress induces cardiac dysfunction and energy depletion through mechanisms independent of circulating cortisol.

CARDIO Cardiac responses to changing environmental conditions

Acclimation to mild static and cyclic hypoxia reduces cardiac thermal tolerance in brook char (Salvelinus fontinalis); potential contribution of novel lactoylated amino acids

MacCormack, Tyson[1] (*tmaccormack@mta.ca*) Oral Presentation
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Abstract: Climate change is increasing the frequency and severity of both diel cycling hypoxia (DCH) and high temperature events in freshwaters. We previously demonstrated that salmonids exposed to DCH have considerable scope to enhance their hypoxia tolerance but trade-offs enabling this were not identified. To address this gap in knowledge, we studied interactions between acute and extended hypoxia and cardiac thermal tolerance in brook char. Fish were exposed to normoxia, diel cycling hypoxia [DCH: 12 h at 40% dissolved oxygen saturation (DO2) overnight], or static mean hypoxia (MH, 70% DO2) for either 2 or 14-days. Fish were sampled directly following exposure or subjected to a modified rapid screening protocol for cardiac thermal tolerance before sampling. Hypoxic conditions induced a temporary fasting response and disruptions in tissue osmotic balance, some that persisted for 14 days. Maximum heart rate and temperature at maximum heart rate decreased in both MH and DCH fish after 14 days, suggesting acclimation to hypoxia, even at mild levels, can decrease cardiac thermal tolerance. Substantial lactate accumulation occurred in fish that completed the cardiac rapid screening protocol. This facilitated the first description of lactoylated amino acid (e.g., “Lac-Phe”) levels in plasma and tissues of a fish, including the identification of a novel compound with potential regulatory importance. Our results demonstrate that enhancing hypoxia tolerance may occur at the expense of thermal tolerance in brook char. We also highlight the potential importance of novel lactoylated amino acids in mediating metabolic and physiological responses to environmental stress in fish.

CARDIO Cardiac responses to changing environmental conditions

Anaerobic Metabolism and Acidosis: Supporting Heart Function in Anoxic Pacific Hagfish (Eptatretus stoutii)

Yutsyschyna, Maria[1] (*myusishe@uoguelph.ca*) Oral Presentation
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Abstract: Pacific hagfish (Eptatretus stoutii) can survive 36 hours of anoxia. Previous research demonstrates that the systemic heart reduces rate and increases stroke volume

to save energy, sustaining itself entirely through glycolysis. This requires managing the delivery of glucose, clearance of lactate, and buffering of metabolic acidosis. To determine how cellular conditions change through time in response to anoxia exposure we measured the concentrations of glucose and lactate in plasma and tissues, along with blood and intracellular pH, during anoxia exposure and reoxygenation. The heart demonstrated strong glycolytic activity with lactate being 36-fold higher in animals exposed to 24 hours of anoxia compared to controls. Plasma glucose increased 6-fold after 30 hours of anoxia as stores are mobilized to support heart function. Lactate levels in the blood were 300-fold higher after 30 hours of anoxia compared to controls. This suggests that the blood acts as a lactate store. These levels started to decline after 6 hours of reoxygenation. Metabolic acidosis was significant with plasma pH declining from a control of 8.1 to 7.0 over 30 hours of anoxia. Recovering to 7.3 after 6 hours of reoxygenation. Intracellular pH was less disrupted with control values of 6.8 – 6.9 across the brain, liver, and muscle. A low of 6.7 was measured in all three tissues after 30 hours of anoxia. In summary, Pacific hagfish tolerate significant lactate loads and acidosis during anoxia with recovery not yet complete after 6 hours. This tolerance both supports and is facilitated by continued heart function during anoxia.

CARDIO Cardiac responses to changing environmental conditions

Swimming Performance of Hagfish in Hypoxia

Shaftoe, Jared (*wjshaftoe@gmail.com*) Oral Presentation
Co-Author: Gillis, Todd
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Abstract: Hagfish are important scavengers and benthic predators in marine ecosystems. They must find food, escape predators, and hunt in water characterized by low oxygen saturation. How hagfish metabolism is affected by these activities is poorly understood so it is important to determine how they survive in hypoxic environments and respond to environmental disturbances. This project focuses on how hagfish with different tolerance to hypoxia meet the energetic demands of swimming in hypoxia. Pacific hagfish (Eptatretus stoutii) and Atlantic hagfish (Myxine glutinosa) were used to compare how anoxia tolerance corresponds to swimming ability. In this study, hagfish were fitted, for the first time, with implantable ECG biologgers to trace heart rate at rest and after exhausting swimming in 100% (normoxia) and 10% (hypoxia) oxygen-saturated seawater. Oxygen consumption of hagfish was not affected up to 7 days post surgery. Maximum voluntary swim speeds of Pacific hagfish were found to be comparable in normoxia and hypoxia. However, the time to exhaustion was 35% shorter for hypoxic Pacific hagfish ($p < 0.05$). Both Pacific and Atlantic hagfish swam more readily in hypoxia than in normoxia. The increase in activity associated with swimming did not result in a significant increase in heart rate. Collectively, these results suggest an increased reliance on anaerobic pathways with little increase in systemic blood flow during exhausting exercise. This work presents novel characterization of the swimming capacity of hagfish in hypoxia. It also provides a method to evaluate the effect of hypoxia tolerance on swimming ability in deep-sea fishes.

CARDIO Cardiac responses to changing environmental conditions

Cardiac Mitochondrial Function and the Temperature-Dependent Physiology of Tropical Marine Fishes

Gamperl, Anthony[1] (*kgamperl@mun.ca*) Poster Presentation
Co-Authors: Clow, Kathy[1]; Syme, Doug[2] and Seamone, Scott[3]
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Abstract: The scaled sardine, Nassau grouper and schoolmaster snapper are tropical marine fishes that differ greatly in their thermal tolerance (CTMax = ~ 34.9, 36 and 38.9oC, respectively) and metabolic capacity (~3000, 400 and 1000 mg O2 kg-1 hr-1, respectively). Differences in heart size, haemotocrit, and myocardial contractile functioncontribute to these inter-specific differences. However, mitochondrial function is also a key determinant of a fish's metabolic capacity and thermal tolerance, and thus, we measured the respiration and reactive oxygen species (ROS) production of isolated ventricular mitochondria from these species at their acclimation temperature (~26oC) and at 34oC. At 26oC, there were no differences in these variables between snapper and sardine. However, while grouper mitochondria had a 2-fold better RCR, they had an even higher (by ~2.5-3.0 fold) ROS production and cytochrome C-induced CI-CII-linked respiration (indicative of lower outer mitochondrial membrane integrity). Raising temperature to 34oC increased State 3 CI-linked respiration in all groups, and resulted in lower values for RCR. However, State 3 CI+CII-linked respiration and RCR in the sardine were only ~ 50% of that measured in the other two species, whereas the difference in cytochromeC-induced CI+CII-linked respiration grew even larger. These data strongly suggest that the respiration of individual mitochondria does not explain the much higher metabolic capacity of sardines, but that: compromised mitochondrial function contributes to the earlier cardiac failure and lower acute thermal tolerance of this species; the grouper may be the most susceptible to long-term increases in ocean temperatures due to their much higher values for ROS production.

CARDIO Cardiac responses to changing environmental conditions

Differential impact of the PAH fluorene and its associated oxyPAHs on the cardiac contractility of juvenile Chinook salmon (Oncorhynchus tshawytscha)

Labonte, Jennessa (*jrlabonte@alaska.edu*) Poster Presentation
Co-Authors: Stecyk, Jonathan; Tomco, Patrick and Redman, Zachary
University of Alaska Anchorage

Abstract: Alaska hosts the most productive Pacific salmon fisheries in the world, with an importance to the economy, recreational use, and subsistence lifestyles. It is well established that fuel spills, which release toxic polycyclic aromatic hydrocarbons (PAHs) into the environment, are detrimental to aquatic organisms. However, a "regulatory blindspot" exists for oxidized polycyclic aromatic hydrocarbons (oxyPAHs), which are

formed from the biological and chemical weathering of parent PAHs, making them more water-soluble, bioavailable, and potentially more toxic. This is especially of concern for Alaskan salmon, where long hours of sunshine in summer increases oxyPAH formation. Given that cardiac performance underlies salmonid survival and reproductive success, we investigated the effect of 3, 30, and 300 µM fluorene (parent PAH), and its associated oxyPAHs, 9-fluorenone, 9-hydroxyfluorene, 2-hydroxy-9-fluorenone, 2,2-dihydroxybiphenyl, and biphenyl-2-carboxylic acid, on maximal contractile force (Fmax) of Chinook salmon juveniles isometrically-contracting, electrically paced (0.2 Hz; N=10-13 per exposure group) ventricular strips. We hypothesized that oxyPAHs would more negatively affect cardiac contractility than fluorene. Indeed, Fmax of strips exposed to fluorene did not statistically significantly differ from that of solvent (DMSO) controls, whereas Fmax decreased (maximal reductions of 17-21% at 300 µM) in strips exposed to the oxyPAHs. Moreover the most water-soluble oxyPAHs, 2,2-dihydroxybiphenyl and biphenyl-2-carboxylic acid, induced a statistically significantly greater depression of Fmax than fluorene. These findings suggest that fuel spill responses at high latitudes may require specific monitoring and mitigation due to the heightened negative effects of oxyPAHs on the cardiac, and by extension, individual performance of salmonids.

CARDIO Cardiac responses to changing environmental conditions

Haematological and biochemical effects of the rapid screening protocol for cardiac thermal tolerance in brook char (Salvelinus fontinalis)

MacCormack, Tyson[1] (*tmaccormack@mta.ca*) Poster Presentation
Co-Authors: Minocha-McKenney, Rajan[1]; Tector, Simon[2]; Pabody, Claire[1] and Lamarre, Simon[3]
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Abstract: Maximum cardiac function plays an important role in supporting acute thermal tolerance in fish. Blood flow measurements require invasive surgical manipulations and specialized training and instrumentation that is not accessible to many researchers. The cardiac rapid screening protocol or Arrhenius Breakpoint Temperature Test is a straightforward, non-terminal electrocardiography method that measures the temperature dependence of maximum heart rate in anesthetized fish. While it offers a variety of advantages over other techniques, little is known about how the rapid screening protocol affects the stress physiology and biochemistry of the animal being studied. As part of a larger project on trade-offs between hypoxia acclimation and thermal tolerance, we measured haematological and biochemical parameters, as well as markers of physiological stress and tissue fluid balance in fish that had completed the cardiac rapid screening protocol. In some fish, lactate levels reached 45 mM in plasma, exceeded 70 mM in heart, and were elevated in all tissues of all fish examined. Tissue water and taurine content were disrupted, suggesting a significant osmotic disturbance. Plasma cortisol levels were massively elevated in all fish, surpassing 500 ng/mL in several cases. Levels of lactoylated amino acids were also higher in multiple tissues. The rapid screening protocol induces a profound physiological and metabolic stress in fish which should be accounted

for in field experiments where animals will be released after testing. Consideration should also be paid to the secondary effects of anaerobic metabolites and stress hormones on the various cardiac endpoints collected in the protocol.

CARDIO Cardiac responses to changing environmental conditions

A field study of interspecific anatomical diversity & comparative scope for seasonal plasticity of the cardiorespiratory organs using 15 species of co-habiting stream fish

Manchester, Elizabeth (*emanches@uoguelph.ca*) Poster Presentation
Co-Authors: Turko, Andy
University of Guelph

Abstract: In their natural habitats, fish are routinely exposed to seasonal fluctuations in abiotic conditions, such as temperature and oxygen availability. One major physiological challenge they must face is the transition between Summer and Winter climates, as many native species remain active year-round. As fish are ectothermic, the transition to Winter can be significantly stressful for metabolism and physiology; ultimately influencing organismal fitness and survival. Tissue plasticity, particularly regarding the cardiorespiratory organs, may aid fish in coping with the challenges associated with harsh winter conditions. Previous research shows that remodeling of the heart and gills is a strategy used by fish to maintain cardiac output and meet oxygen demands during prolonged exposure to challenging conditions. However, the capacity for such for plasticity in many native species, as well as how plasticity manifests uniquely in multiple species within a shared ecosystem, remains unknown. In the current study, we collected heart and gill tissues from 15 wild-caught stream species in both the Summer and Winter seasons to empirically assess the scope for seasonal tissue remodeling in the wild. This comparative approach will place species on a “plasticity continuum” to explore how plasticity may relate to environmental sensitivity. Interspecific differences in gross cardiac and gill anatomy will also be explored. These species occupy different micro-niches within the same stream, offering a unique opportunity to connect cardiorespiratory form and ecological function. Shared evolutionary history between species allows us to explore evolutionary influences on cardiorespiratory traits within a shared environment.

CARDIO Cardiac responses to changing environmental conditions

Thermal acclimation potential of juvenile Alaskan Chinook (Oncorhynchus tshawytscha)

Wagner-Jones, Amelia (*alwagnerjones@alaska.edu*) Poster Presentation
Co-Authors: Williams, Emily; Snyder, Augustus and Gilbert, Matthew
University of Alaska Fairbanks

Abstract: Northern climates are defined by extreme seasonality and these environmental challenges influence the functioning of Northern fish species, which have substantial interspecies variation in how they address these challenges and intraspecies variability

with local climate. As ectotherms, fish body temperature and thus heart rate increases proportionally with environmental temperature, and some species display substantial seasonal plasticity to compensate for this innate effect. While seasonal plasticity of cardiac function has been explored in a variety of northern fish species and lower latitude Chinook strains, this topic of research has not been explored in northern Chinook populations, which may encounter water temperatures ranging from ~0°C in the winter to ~13°C in the summer, with periodic peaks >21°C. Our study examined how thermal acclimation alters cardiac function and thermal sensitivity of Yukon River juvenile Chinook, and the resulting effects on function under acute thermal stress. This was achieved using an electrocardiogram approach with induction of maximum heart rate and assessment of cardiac metabolism of juvenile Chinook acclimated to five temperatures (12.5, 15, 17.5, 20, and 22.5°C). Our results suggest that these fish show substantial thermal acclimation plasticity, with warm-acclimated fish showing a marked resetting of their heart rates and an increase in their peak performance and upper thermal limits of whole animal and cardiac function. Additionally, assessment of cardiac tissue biochemistry shows significant thermal compensation occurring in both cold and warm acclimated fish to offset the effects of temperature on metabolic function.

DEEP BLUE The deep blue sea: Probing the physiology and behaviour of large marine fishes

The deep blue sea: Probing the physiology and behaviour of large marine fishes
McKenzie, David and Holly Shiels

Abstract: The physiology and behaviour of large marine fishes, such as tunas, billfishes, and sharks, has interested fish biologists for over a century, from both fundamental and applied fishery perspectives. They often inhabit and migrate through vast open oceans, which makes direct observation, capture, and tracking challenging. The stress of capturing and handling large fishes can have significant impacts on their physiological state, and they are difficult to house in laboratories for controlled studies. As a consequence, most large marine fish species are poorly studied and data deficient. Nonetheless, the physiology and behaviour of some large marine fishes are beginning to be understood and this session will provide a forum to present the latest advances in knowledge. We welcome talks from the biologging and biotelemetry community; on behavioural studies including of group living, or on research that infers aspects of whole animal physiology and behaviour from controlled studies on organs, tissues, and molecular processes. We dedicate this session to the memory of John F. Steffensen, who had a passion for pelagic fishes and spent many happy years probing their physiology and behaviour.

DEEP BLUE The deep blue sea: Probing the physiology and behaviour of large marine fishes
Tracking the Physiology Pelagic Fish From the lab to the Blue Serengeti
Block, Barbara - Oral Presentation

Abstract: Large marine predators (tunas, marlins and sharks) are important top consumers in marine ecosystems and currently face unprecedented challenges and uncertain futures. Climate variability, overexploitation and rapidly changing ecosystems impact their abundance, foraging ecology, spawning and distributions across large ocean ecosystems. Large marine predators are important top consumers and their depletions can have cascading effects on lower trophic levels in both coastal and open-ocean waters. Improved understanding of their physiology, niche preferences, evolutionary constraints and physiology in these changing ocean ecosystems are critical for preventing extinctions, loss of biodiversity and disruption of ecosystem services. I will overview how we have combined lab and field physiology, with the objective focused on integration of lab measurements with electronic tagging data (biologging) acquired at sea on wild fish. This provides the capacity to observe, model and predict the movements, physiology, habitat use and trans-oceanic migrations of pelagic fish in relationship to metabolic, cardiac and oceanic processes across a large range of ecological scales. Tagging enables exploration of how and why fish choose to be residents in coastal seas versus their capacity for long trans-oceanic migrations. Tagging now provides physiology on thermoregulation, cardiac physiology, energy consumption and use of open ocean habitats from the surface to the

deep sea. Biologging enables fish researchers to monitor pelagic fish biology at unprecedented scales, which has the capacity to improve international fisheries management. Investments in reducing the size of biologging instruments, increasing the bandwidth of data communications are improving our capacity to track the physiology of pelagic fishes in changing seas.

DEEP BLUE The deep blue sea: Probing the physiology and behaviour of large marine fishes

Natural isotope tracer-based estimation of field metabolic rates of wild pelagic fishes: approaches, applications and challenges

Trueman, Clive (*trueman@soton.ac.uk*) Oral Presentation
University of Southampton

Abstract: Large pelagic fish species present numerous practical challenges for physiologists, resulting in a relative dearth of physiological data which is particularly concerning given the important ecological, socioeconomic and cultural roles fulfilled by large pelagic species. Biologging and chamber respirometry are the main sources of physiological data for large pelagic species. Natural stable isotope-based tracers in biomineralised carbonate-bearing tissues such as otoliths provide a retrospective record of total energy use (i.e. field metabolic rate) which can be applied to (almost) any wild living marine teleost and potentially elasmobranchs. Here I will first outline the mechanism underpinning stable-isotope based inference of FMR. Otolith-inferred estimates of time-averaged chronic FMR measured to date in wild pelagic teleosts range from c.10 mgO2kg-1h-1 in non-migratory deep pelagic fishes to >450 mgO2kg-1h-1 in large tunas. Complimentary isotopic proxies for experienced temperature and FMR derived from the same otolith samples allow generation of thermal performance curves and realised thermal sensitivity of FMR. Using these data, we identify temperature conditions imposing physiological limits on juvenile Atlantic bluefin tuna in natural conditions, demonstrate differential physiological responses to warming among large pelagic fish juveniles in the central Mediterranean Sea, and compare realised metabolic rates between wild and ranched tuna. While otolith-based isotopic proxies are relatively well studied, potential exists to also recover field physiological information from carbonate-containing bioapatite (e.g. bone and tooth mineral) in elasmobranchs. Preliminary data supports isotopic theory suggesting that physiological information should be recoded in these structures, but with considerable complications.

DEEP BLUE The deep blue sea: Probing the physiology and behaviour of large marine fishes

Effects of Body Size on Spontaneous Swimming Activity and Energetics in Atlantic Bluefin Tuna

Thambithurai, Davide[1,2,3] (*davide.thambithurai@gmail.com*) Oral Presentation
Co-Authors: Cortese, Daphne[1,2,3]; Steffensen, John F.[4]; Reglero, Patricia[5]; Ortega, Aurelio[5]; de la Gándara, Fernando[5]; Rouyer, Tristan[1,2,3] and McKenzie, David J.[1,2,3]
[1]MARBEC, [2]University of Montpellier, [3]CNRS, [4]University of Copenhagen, [5]Spanish Institute of Oceanography

Abstract: Movement and locomotor efficiency are central to fitness in fish, yet the energetic consequences of swimming remain poorly understood in many species. Atlantic bluefin tuna (ABFT, Thunnus thynnus) undertake extensive basin-scale migrations, growing from a few millimetres as larvae to over three metres as adults while experiencing highly variable environmental conditions. In this study, we investigated how body size influences spontaneous swimming speed (Uspo) and locomotor efficiency, quantified as cost of transport (COT), and examined the effects of environmental oxygen on these traits. We tested three hypotheses: (1) Uspo scales allometrically with body size; (2) swimming efficiency varies with size; and (3) Uspo and COT are influenced by oxygen availability.
We measured Uspo across four size classes of ABFT in large tanks and compiled metabolic data to estimate COT across a >1000-fold range in body mass. As expected, absolute swimming speed increased with size, whereas relative swimming speed (body lengths s^{-1}) decreased. Larger fish exhibited substantially lower COT, indicating greater locomotor efficiency. Oxygen concentration showed weak evidence of affecting Uspo and no conclusive effect on COT under hyperoxic versus normoxic conditions.
Our results reveal strong allometric scaling of swimming activity and energy use in ABFT, with smaller individuals incurring proportionally higher energetic costs, and environmental oxygen having limited effects under the conditions tested. These findings provide critical data for mechanistic models, enhancing our understanding of how body size and environmental factors shape locomotor performance in this highly migratory species.

DEEP BLUE The deep blue sea: Probing the physiology and behaviour of large marine fishes

Atlantic bluefin tuna recovery from catch-and-release

Birnie-Gauvin, Kim (*kbir@aqua.dtu.dk*) Oral Presentation
Co-Authors: Pedaccini, Marie
Technical University of Denmark

Abstract: Atlantic bluefin tuna (ABFT) are among the most sought after fish, both for harvest and for recreational angling. Catch-and-release is common for ABFT caught by recreational anglers, with the assumption that the fish survive. However, relatively little is known about the duration of their recovery following release from capture, and the factors that affect this recovery. To address this knowledge gap, we captured 48 ABFT via rod-and-reel, and equipped them with either 1) a biologging package containing loggers for acceleration, depth and temperature, or 2) a pop-up satellite archival tag (PSAT). The fish were also

sampled for blood and muscle to assess metabolites, and their potential link to recovery. We also explored other factors that may affect recovery, such as fight time, and time on deck. We found that ABFT were more active following release, and showed marked differences in their diving behaviour immediately following release and during recovery than what is considered their typical behaviour.

DEEP BLUE The deep blue sea: Probing the physiology and behaviour of large marine fishes

Mesothermic fishes face high fuel demands and overheating risk in warming oceans

Payne, Nicholas[1] (*paynen@tcd.ie*) Oral Presentation
Co-Authors: Jackson, Andrew[1]; Snelling, Edward[2]; Peralta-Maraver, Ignacio[3]; Cade, David[4]; Sims, David[5]; Watanabe, Yuuki[6]; Chapple, Taylor[7]; Mcinturf, Alexandra[7]; Dolton, Haley[1]; Da Costa, Ivo[8]; Queiroz, Nuno[8]; Goldbogen, Jeremy[4]; Sousa, Lara[8]
[1]Trinity College Dublin; [2]University of Pretoria; [3]University of Granada; [4]Stanford University [5]Marine Biological Association of UK; [6]SOKENDAL, [7]Oregon State University, [8]University of Porto

Abstract: Body size and temperature set metabolic rates and the pace of life, yet our understanding of the energetics of large fishes is uncertain, especially of warm-bodied mesotherms, which can heavily influence marine food webs. We developed an approach to estimate metabolic heat production in fishes via biologging, then revealed how routine energy expenditure scales with size and temperature from 1-mg larvae up to 3-tonne mega-planktivorous sharks. We found mesotherms use ~ 4 times more energy than ectotherms and identified a scaling mismatch whereby rates of heat production increase faster than heat loss as body size increases, with larger fish becoming increasingly warm bodied. This scaling imbalance creates an overheating predicament for large mesotherms, helping explain their cooler biogeographies. Contemporary mesotherms face high fuel demands and overheating risks; a concern given their disproportionate demise during prior climate shifts

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DEEP BLUE The deep blue sea: Probing the physiology and behaviour of large marine fishes

Fine-scale spawning dynamics of Atlantic bluefin tuna

Cortese, Daphne[1,2,3] (*daph.cortese@gmail.com*) Oral Presentation
Co-Authors: Deguara, Simeon[4]; Galea, Justin[4]; Thambithurai, Davide[5]; Bernard, Serge[6,2,3]; Kerzerho, Vincent[6,2,3]; Cosnard, Nolwenn[7]; Wendling, Bertand[7]; Derridj, Olivier[1,2,3]; Bjarnason, Asgeir[8]; Steffensen, John F[9]; McKenzie, David J.[1,2,3,] and Rouyer, Tristan[1,2,3]
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Abstract: The Atlantic bluefin tuna (Thunnus thynnus) is a large pelagic predator with physiological specializations for long-distance migrations between feeding and spawning grounds, including partial endothermy and obligate ram ventilation.However, despite its large-scale migration movement being well documented, the fine-scale dynamics of

spawning behaviour remain poorly understood. We monitored 24 individuals (mass range: 25 to 200 kg) over one year in an offshore sea-pen off Malta using high-resolution acoustic telemetry (n = 24) and heart rate biologgers (n = 17). We detected recurrent nocturnal episodes of elevated swimming and cardiac activity during the summer spawning period. By combining behavioural and physiological data, we identified distinct spawning-associated signatures, providing the first high-resolution description of the spawning behaviour in this species. Overall, this study offers novel new mechanistic insight into bluefin tuna ecology and tools to better identify spawning behaviours in wild populations.

DEEP BLUE The deep blue sea: Probing the physiology and behaviour of large marine fishes

Effects of seasonal temperatures on cardiac and swimming activity in Atlantic bluefin tuna

McKenzie, David J.[1,2,3] (*david.mckenzie@cnrs.fr*) Oral Presentation
Co-Authors: Cortese, Daphne[1,2,3]; Deguara, Simeon[4]; Galea, Justin[4]; Thambithurai, Davide[5]; Derridj, Olivier[1,2,3]; Kerzerho, Vincent[6,2,3]; Steffensen, John F.[7]; Bjarnason, Asgeir[8]; Cosnard, Nolwenn[9]; Wendling, Bertand[9]; Bernard, Serge[6,2,3] and Rouyer, Tristan[1,2,3]
[1]MARBEC, [2]University of Montpellier, [3]CNRS, [4]AquaBioTech Group, [5]University of Glasgow, [6]LIRMM, [7]University of Copenhagen, [8]Star-Oddi, [9]Sathoan

Abstract: The Atlantic bluefin tuna Thunnus thynnus (ABFT) is a large apex predator with physiological adaptations for long-distance oceanic migrations.While their heart is ectothermic and functions at ambient temperatures, aerobic swimming muscles retain metabolic heat to maintain performance in cold waters, and ABFT must swim ceaselessly to ram-ventilate.We studied effects of seasonal acclimatisation on cardiac and locomotor activity in ABFT (mass range 25 to 200 kg) in a sea-pen over one year, that experienced temperatures from 15°C in winter to 28°C during a summer heatwave. Biologgers (n=17) recorded heart rate (fH) and temperature every 12 minutes throughout; high-residency acoustic telemetry (n=24) recorded swimming activity every 2 seconds for 24 hours each fourth day. To explore thermal sensitivity of expressed scope for performance, we binned each individual's fH and swimming speed (U) data into 1°C intervals, and took the 10th quantiles (10-Q) and 90th quantiles (90-Q) as indexes of essential basal activity and spontaneous maximum activity, respectively, and the net difference as indicator of scope.The 10-Q for fH increased across the temperature range with a Q10 of ~2; the 90-Q increased from 15 to 27°C but declined significantly at 28°C, with expressed scope for fH greatest between 25 and 27°C.For U, the 10-Q and 90-Q showed some thermal sensitivity but overall effects were modest, and spontaneous scope for Udid not vary consistently with temperature.Such descriptive exploratory data can be used to improve mechanistic models for effects of temperature on ABFT performance and migration.

DEEP BLUE The deep blue sea: Probing the physiology and behaviour of large marine fishes

Body size relationships in cardiac morphology and electrocardiographic intervals of Atlantic Bluefin tuna (ABFT)

Bjarnason, Asgeir[1] *(asgeir@star-oddi.com*) Oral Presentation
Co-Authors: Cortese, Daphne[2]; Deguara, Simeon[3]; Galea, Justin[3]; Thambithurai, Davide[4]; Bernard, Serge[5,6,7]; Kerzerho, Vincent[5,6,7]; Wendling, Bertand[8]; Derridj, Olivier[9,6,7]; McKenzie, David J.[9,6,7] and Rouyer, Tristan[9,6,7]
[1]Star-Oddi; [2]Ifremer; [3]AquaBioTech Group, [4]University of Glasgow, [5]LIRMM, [6]University of Montpellier, [7]CNRS, [8] Sathoan, [9]MARBEC

Abstract: Abstract Text: Understanding how cardiac morphology and electrical activity scale with body size is essential for interpreting how the cardiovascular system supports the high metabolic demands of large pelagic fishes. Atlantic bluefin tuna (Thunnus thynnus) are among the largest and most metabolically active teleosts, yet relatively little is known about how cardiac morphology and electrophysiological properties change across body sizes. In this study, we investigated relationships among body mass, ventricular mass, heart rate, and electrocardiographic (ECG) parameters (QRS duration, QT interval, and PR interval) in eight bluefin tuna spanning body masses from 25 to 94 kg. Measurements of cardiac morphology, heart rate, and electrical conduction intervals were obtained for each individual at two temperatures (~16 °C and ~26 °C), with analyses restricted to fasting periods to minimize any influence of specific dynamic action of food. Preliminary results show that ventricular mass scaled positively with body mass (R^2 = 0.49). Similarly, QRS duration increased with body mass at both temperatures (R^2 = 0.60 at 16 °C and R^2 = 0.47 at 26 °C), suggesting that ventricular depolarization time increases with cardiac size. In contrast, heart rate , PR interval, and QT interval showed little or no correlation with body mass. These findings suggest that, while cardiac morphology and ventricular electrical conduction scale with body size in the Atlantic bluefin tuna, other aspects of cardiac electrophysiology may be relatively size-independent, highlighting potential adaptations that maintain cardiac performance across a wide range of body sizes in this highly active species.

DEEP BLUE The deep blue sea: Probing the physiology and behaviour of large marine fishes

Thermal sensitivity of cardiac function in the Greenland Shark

Shiels, Holly[1] (*holly.shiels@manchester.ac.uk*) Oral Presentation
Co-Authors: McKenzie, David J.[2]; Brill, Rich[3]; Bernal, Diego[4]; Andersen, Lars Emil[5]; Vedel, Kristian[5]; Steffensen, Kirstine Fleng[5]; Bushnell, Peter[6] and Steffensen, John F[5]
[1]University of Manchester; [2]MARBEC, CNRS; [3]Virginia Institute of Marine Sciences; [4]University of Massachusetts Dartmouth; [5]University of Copenhagen, [6]Indiana University South Bend

Abstract: Greenland sharks (Somniosus microcephalus) occupy a narrow thermal niche. They live in the cold deep waters of the Arctic and sub-Arctic and where water at depth rarely exceeds 5C. As global ocean temperatures warm, cold stenotherms are particularly vulnerable to the direct effects of temperature on physiological processes. The heart is a

linchpin in survival during thermal stress and important for matching oxygen supply with metabolic demand. Here we examined the thermal sensitivity of the Greenland shark heart to understand its vulnerability to a warming climate. In vivo heart rate assessed via ECGs in anaesthetised force-ventilated animals showed extreme thermal sensitivity. However, exogenously paced isolated ventricular muscle strip preparations were able to develop force at temperatures approaching 20C, much higher than the whole organism thermal tolerance. This could indicate a failure of electrical propagation at warm temperatures in vivo. This was probed by mapping the effect of temperature on conduction velocity. These findings are discussed in relation to satellite tagging data showing thermal distribution in the wild.

DIGESTION The fish gut: Digestion and beyond

The fish gut: Digestion and beyond

Patricia Ferreira and Jonathan Wilson

Abstract: The fish gut is a multifunctional organ system with roles extending far beyond its immediate digestive and absorptive processes. It is a major endocrine organ in vertebrates, and plays fundamental roles in the crosstalk with other systems, notably the brain. The gut microbiome is also emerging as a key piece in this regulatory puzzle. Moreover the gut organs have evolved different adaptations and forms throughout the evolution of vertebrates, with particular diversity being present in fish. How this diversity has been shaped to respond to selective pressures and helped fish achieve efficient digestion and energy homeostasis has been, and still is a hot topic in fish research. This symposium aims to bring together researchers from different areas of gut research to present their recent research findings from the molecular to organismal level, bridging knowledge gaps and inspiring the exchange of ideas on the role of the different aspects of gut physiology in applied (aquaculture), ecological and evolutionary contexts.

DIGESTION The fish gut: Digestion and beyond

Genomic foundations of the vertebrate stomach

Castro, Filipe[1] *(filipe.castro@ciimar.up.pt)* Oral Presentation
Co-Authors: Teixeira, João[2]; Ferreira, Patricia[3]; Wilson, Jonathan[4]
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Abstract: Vertebrate evolution has been punctuated by the emergence and elaboration of organs that enabled major ecological transitions and the exploitation of new niches. Among these innovations, the stomach represents a key digestive organ defined by the evolution of acid–peptic digestion. By coupling hydrochloric acid secretion with the activity of proteolytic enzymes such as pepsin, the stomach expanded the capacity of vertebrates to process protein-rich diets, improve nutrient extraction, and exploit a wider range of trophic strategies. Despite its central role in vertebrate physiology, fundamental questions remain. How did the stomach originate during vertebrate evolution? Which genomic innovations enabled the emergence of acid secretion and peptic digestion in the ancestor of gnathostomes? And is the molecular architecture underlying stomach function conserved across extant vertebrate lineages? Intriguingly, the stomach has also been repeatedly lost during vertebrate evolution, particularly among several teleost fish lineages, raising questions about the evolutionary flexibility of digestive systems and the selective forces that permit the disappearance of such a seemingly fundamental organ. In this talk, I explore the evolutionary origins, diversification, and recurrent loss of the vertebrate stomach by integrating comparative genomics, gene repertoire evolution, and physiological insights across vertebrate taxa. This framework provides a molecular perspective on how complex organs emerge, diversify, and sometimes disappear during evolution.

DIGESTION The fish gut: Digestion and beyond

Food for thought: The role of serotonin in mediating energy metabolism via the gut

Gomes Ferreira, Patricia (*patricia.ferreira@uottawa.ca*) Oral Presentation
Co-Authors: Scott, Brenna; Tea, Michael; Gilmour, Kathleen
University of Ottawa

Abstract: The gastrointestinal tract is a major endocrine organ that coordinates digestion, metabolism, and feeding behavior through bidirectional gut–brain communication. Although serotonin (5-hydroxytryptamine; 5-HT) was originally identified as a gut hormone, research has largely focused on its neuro-modulatory roles in behavior. Renewed interest in peripheral serotonin has revealed key functions in mammalian metabolic regulation and lipid and glucose homeostasis. Circulating 5-HT levels are regulated by reuptake via the serotonin transporter (Sert; slc6a4). Unlike mammals, zebrafish have two Sert paralogues, slc6a4a and slc6a4b, providing a unique opportunity to investigate functional specialization of serotonergic regulation in metabolic tissues. Here, we examined the role of gut serotonin in zebrafish using knockout models lacking slc6a4a and/or slc6a4b. We characterized postprandial regulation of Sert paralogues in the intestine and assessed the metabolic consequences of disrupted serotonin reuptake. Preliminary findings revealed genotype-specific alterations in appetite and lipid storage. Notably, we demonstrated that Sert paralogues are postprandially regulated in the gut and investigated the expression of candidate genes linked to carbohydrate and lipid metabolism in both the intestine and the liver. These findings will contribute to further establishing zebrafish as a system to dissect peripheral serotonergic control of metabolism and provide new insight into gut-derived mechanisms linking serotonin signaling with metabolic regulation.

DIGESTION The fish gut: Digestion and beyond

Structural support or functional necessity: Evaluating the importance of the gastric proton pump β-subunit in gastric function using gene knockout.

Seguin, Aiden[1] (*segu4400@mylaurier.ca*) Oral Presentation
Co-Authors: Ferreira, Patricia[2]; Wilson, Jonathan[1]
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Abstract: The vertebrate stomach, characterized by acid-peptic digestion, is a specialized evolutionaryadaptationenhancingdigestion.Acidsecretionis dependent on the heterodimeric gastric proton pump (H+/K+-ATPase; HKA), encoded by the genesatp4aandatp4b. Atp4a is the catalytic subunit, andAtp4bhas roles in membrane targetingand complex stability. The emergence of the stomach led to an extension of available protein sources, and an array of advantages including immune defense and improved cell lysis. Despite the advantages associated with a gastric phenotype, the dynamics of the two HKA subunitsremainto be fullyunderstood,especially in the contextofthe secondary loss of gastric acidificationobservedin a quarter ofteleostspecies. We have previouslyestablishedanatp4aknockout inAstyanax mexicanusand described the phenotype, focused on metabolism,modulation ofatp4bexpression,and HKA membrane

trafficking. In this study, wehavegeneratedanatp4bcrispant(rapid, F0 generation, somaticknockout)inA.mexicanususing CRISPR-Cas9 gene editingto investigate therollofAtp4bingastric physiology, cell dynamics and, particularly, the modulation of the expression and membrane recruitment ofAtp4a.Atp4bcrispantsexhibited nodifferences in overallgrowth; however,theydisplayed altered metabolic processes due to a significantreduction in gastric acid secretion.Immunohistochemistryrevealed thatAtp4aandAtp4bboth underwentadownregulation in gastric glands, highlighting the role ofAtp4bin HKA membrane targeting.Ultimately, thisproject advances our understanding of the physiology of gastric secretion and provides insights into the drivers and consequences underlying stomach loss infishes.

DIGESTION The fish gut: Digestion and beyond

Thermal plasticity of intestinal function in satiated and starved European seabass

Ruhr, Ilan[1] (*ilanruhr@gmail.com*) Oral Presentation
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Abstract: The vertebrate gut is a highly plastic organ system, yet the response of the fish intestine to environmental stressors, like temperature perturbations, is poorly studied. Identifying the mechanisms underlying intestinal phenotypic plasticity in fish is critically needed in the era of global warming. As aquatic ecosystems warm, wild fish will need to traverse ever-steeper thermoclines and cultured fish will be confined to ever-hotter waters, potentially leading to acute-warming stresses. In marine teleost fish, rising temperatures elevate oxygen consumption, which is compensated by increasing cardiac output and branchial ventilation, but also by likely boosting intestinal water and ion absorption, the latter being the focus of this study. We investigated intestinal thermal and food-rationing plasticity in European seabass (Dicentrarchuslabrax), after 12 weeks' exposure to 18°C (cold) or 26°C (warm) and feeding 3% body weight 1x or 3x weekly. Using intestinal sacs and Ussing chambers, we subjected the anterior intestine of warm fish to acute cooling (18°C) and cold fish to acute warming (26°C). Acute cooling reduced water/ion absorption in the intestine of satiated and starved warm fish, which was more prominent in starved fish, corresponding to lower metabolic demands associated with lower food uptake and colder water. Conversely, acute warming increased water/ion absorption for satiated cold fish, but had no effect for starved cold fish (partly due to higher tissue conductance; supported by histology and immunohistochemistry). This is a seemingly counterproductive response for starved cold fish, given the increased metabolic demands evoked by acute warming that necessitate higher rates of intestinal water absorption.

DIGESTION The fish gut: Digestion and beyond

Convergent gene losses and pseudogenizations in stomachless fishes

Kato, Akira[1] (*akirkato@life.isct.ac.jp*) Oral Presentation
Co-Authors: Pipil, Supriya[2]; Ota, Chihiro[1]; Kusakabe, Makoto[3]; Watanabe, Taro[2]; Nagashima, Ayumi[1]; Chen, An-Ping[4]; Islam, Zinia[5]; Hayashi, Naoko[5]; Wong, Marty Kwok-Shing[6]; Komada, Masayuki[1]; Romero, Michael F.[4] and Takei, Yoshio[2]
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Abstract: Regressive evolution in independent lineages often shows convergent phenotypes; however, little is known about their respective genotypic convergences. Several groups of teleost fishes display secondary loss of the stomach as a model of regressive evolution, and four major gastric genes, atp4a, atp4b, pgc, and pga2 have been shown to be co-deleted in agastric (stomachless) fishes. To analyze genotypic convergence among agastric fishes in more detail, we investigated common gene losses using genome databases of these fishes (e.g., zebrafish, medaka, pufferfish, and wrasse) and compared them to those of gastric fishes (e.g., gar, catfish, trout, stickleback, tilapia, and cod). Four genes, slc26a9, kcne2, cldn18.1, and vsig1, were newly found to be co-deleted or pseudogenized in most of agastric fishes, and kcne2 and vsig1 was also absent or pseudogenized in agastric monotreme echidna and platypus, respectively. In stickleback, slc26a9, kcne2, cldn18.1, and vsig1, as well as atp4a, atp4b, pgc, and pga2 were highly expressed in the stomach. In situ hybridization showed that atp4a, atp4b, pgc, pga2, slc26a9, and kcne2 were expressed in gastric gland cells, whereas cldn18.1 and vsig1 were expressed in surface epithelial cells. The cldn18.2, a paralog of cldn18.1, was retained in some agastric fishes and a showed an increased non-synonymous substitution rate when compared with gastric species, suggesting functional constraint relaxation of cldn18.2. These results clarified the novel convergent gene losses at multiple loci among four major groups of agastric fishes, as well as a single gene loss in the echidna and platypus.

DIGESTION The fish gut: Digestion and beyond

Digestive enzyme activities in prickleback fishes (Stichaeidae) appear to be driven by gene expression

German, Donovan (*dgerman@uci.edu*) Oral Presentation
University of California Irvine

Abstract: Digestion is a chemical process and digestive enzymes are the agents of digestion. Researchers have long documented that fishes with different natural diets have varying activity levels of digestive enzymes: herbivores/omnivores have elevated amylase activity, whereas some carnivores have elevated aminopeptidase and/or trypsin activities. Recent work suggests diet-related variation in lipolytic and gastric (pepsin, chitinase) digestive enzyme activities as well. Here, I show how changes in gene copy number (amylase, lipase) or gene expression unrelated to copy number can largely explain these

differences in digestive enzyme activities among closely related prickleback fish species with different diets. Moreover, there are clearly varying degrees of plasticity in digestive enzyme expression and activity in response to laboratory dietary perturbations in species with different diets, with herbivores being less flexible than other species. Thus, at least in prickleback fishes, digestive specialization is largely about gene expression that can include gene copy number variation, or not. Explorations in other fish families that have closely related species with different diets can help explain whether this is a wide-spread phenomenon for digestive enzymes, or whether more specific changes to amino acid sequences might explain enzymatic activities in some species.

DIGESTION The fish gut: Digestion and beyond

Integrating transcriptomics, qPCR and electrophysiology to assess intestinal adaptation to plant-based diets in Greater Amberjack

Fuentes, Juan[1] (*xoan.fuentes@csic.es*) Oral Presentation
Co-Author: Fonseca, Filomena[1]; Agostinho Coelho, Ana Sofia[1]; Martos Sitcha, Juan Antonio[2]
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Abstract: The transition toward more sustainable aquafeeds requires reducing reliance on fishmeal and fish oil through incorporation of plant-based ingredients. While environmentally desirable, this strategy may affect intestinal physiology in fast-growing carnivorous species. This study characterized molecular and functional responses of the anterior intestine of juvenile greater amberjack (Seriola dumerili) to plant-based diets and identified candidate molecular markers associated with epithelial integrity, nutrient absorption, and immune function. Juveniles were fed a commercial control diet or one of two experimental diets in which 50% of animal protein was replaced by plant-derived ingredients differing in wheat flour and wheat gluten content. Anterior intestinal samples were analyzed using RNA sequencing (RNA-seq) to identify diet-responsive genes, followed by functional annotation and validation of selected targets by quantitative PCR (qPCR). In parallel, electrophysiological measurements assessed epithelial transport properties. Principal Component Analysis revealed clear separation between control and plant-based diet groups, indicating diet formulation as the primary driver of transcriptomic variation. Differential expression analyses identified coordinated modulation of genes involved in protein turnover, membrane organization, ion transport, metabolism, and signal transduction. qPCR validation confirmed the reliability of transcriptomic patterns, while electrophysiology revealed diet-associated changes in epithelial transport consistent with molecular responses. Together, these results show that partial fishmeal replacement induces coordinated transcriptional and functional remodeling of the anterior intestine inS. dumerili, highlighting candidate molecular markers of intestinal adaptation to sustainable aquafeeds.

DIGESTION The fish gut: Digestion and beyond

Secondary bile acids impact rainbow trout homeostasis through modulation of feeding and metabolism

Soengas, José L.[1] (*jsoengas@uvigo.gal*) Oral Presentation
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Abstract: Secondary bile acids (SBAs), long considered simple microbial metabolites, are now recognized in mammals as signaling molecules involved in the regulation of feeding behavior and energy metabolism, yet their physiological role in fish remains poorly understood. In this study, we investigated whether SBAs contribute to metabolic homeostasis in rainbow trout (Oncorhynchus mykiss) by modulating feeding and intermediary metabolism. Using intragastric administration of lithocholic acid (LCA), deoxycholic acid (DCA), and their taurine-conjugated forms (T-LCA and T-DCA), we evaluated their intestinal reabsorption, tissue distribution, and regulatory effects on bile acid signaling pathways, feeding regulation and energy balance. We analyzed the expression of bile acid transporters and receptors in the brain, gastrointestinal tract, liver, and white adipose tissue, together with key genes, enzyme activities, and metabolites involved in glucose and lipid metabolism, and assessed short-term feed intake responses. Our results show that SBAs are efficiently reabsorbed and associated with changes in peripheral and central tissues in rainbow trout, where only some of the SBA tested potentially exert specific regulatory effects on FXR and TGR5 signaling. These responses were associated with a slight increase in feed intake and coordinated changes in metabolic pathways in liver and adipose tissue, with unconjugated bile acids showing stronger effects than taurine-conjugated forms. Overall, our findings identify SBAs as novel endocrine regulators of feeding and metabolic homeostasis in teleost fish, revealing a conserved role for bile acid signaling beyond digestion and highlighting its relevance for fish physiology and aquaculture research

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DIGESTION The fish gut: Digestion and beyond

Gastrointestinal nitrogen metabolism in a voracious predator: studying one or studying millions?

Wood, Chris (*woodcm@zoology.ubc.ca*) Oral Presentation
The University of British Columbia

Abstract: The marine staghorn sculpin Leptococcus arrmatus is an excellent model for studying the physiology of feeding. It will voluntarily eat and then slowly digest a massive meal despite being catheterized and confined in a respirometry chamber. A previous study found that during the period of Specific Dynamic Action after the meal, the Nitrogen Quotient, the excretion rate of N- waste (ammonia + urea-N) relative to the consumption rate of oxygen, far exceeded the maximum predicted by standard metabolic theory. The present study tested the hypothesis that this was due to anaerobic production of ammonia

and urea-N without the consumption of oxygen by the millions of microbes in the gut. Fasted sculpins were fitted with rectal catheters and fed a single meal of natural food (consumed ration = 16% body weight of anchovies).Whole animal oxygen consumption, N-waste excretion to the water and N -waste efflux via the anus were monitored for 1 pre-feeding day, followed by 3 post-feeding days, relative to sham-fed control animals. In a second series of experiments, gastro-intestinal ammonia and urea-N production and absorption rates were measured during the 24-to 36-h post-feeding period using novel gut sac preparations. These were filled with their own endogenous chyme and incubated in vitro for 3 h. The results support the hypothesis, suggesting that approximately half of the post-feeding N-waste production occurs in the gut, most of which is absorbed into the body and excreted through the gills, rather than effluxed in the rectal fluid (NSERC Discovery).

DIGESTION The fish gut: Digestion and beyond

Multigenerational gut-brain axis remodeling in marine medaka under chronic hypercapnia

Liu, Tzu-Yen[1] (*rw6526@gmail.com*) Oral Presentation
Co-Authors: Tseng, Yung-Che[2] and Ming-Tsung[1]
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Ocean acidification can disrupt neural function and behavior in marine fish, yethe role of the gut microbiome in mediating multigenerational responses remains unclear. We reared Oryzias melastigma under elevated CO2–induecd hypercapnia (pH 7.6) for three consecutive generations (F0, F1, F2) and examined gut microbiome profiling, brain transcriptomics, and anxiety-related behavior. While alpha diversity remained stable, beta diversity and the Firmicutes/Bacteroidota ratio shifted, indicating altered microbial metabolic potential. F0 displayed acute dysbiosis characterized by increased aerobic chemoheterotrophy and ureolysis, alongside significant downregulation of brain GABA-A receptor activity and increased anxiety-like behaviors. In F1, a distinct metabolic shift toward anaerobic respiratory pathways (e.g., arsenate respiration) coincided with the peak upregulation of brain monocarboxylic acid transport, signaling a critical stage of neuro-microbial realignment. Notably, while compensatory GABAergic signaling persisted into F2, LEfSe analysis identified no specific biomarkers for the F2 microbiota, suggesting that the microbial community reached a new functional equilibrium. Although anxiety-like behaviors persisted, the stabilization of microbial communities and neural pathways suggests that intergenerational tuning of the MGB axis may buffer neurological impairments. These results indicate that chronic CO2–induecd hypercapnia induces coordinated but incomplete adjustment of the gut-brain axis across generations, with microbial community stabilization preceding but not fully restoring behavioral homeostasis.

DIGESTION The fish gut: Digestion and beyond

Specific dynamic action and other related inconveniences

Goodrich, Harriet (*harriet.goodrich@utas.edu.au*) Oral Presentation
Institute for Marine and Antarctic Studies

Abstract: Specific dynamic action (SDA) is often framed as the energetic cost of digestion. In practice, it is far less convenient. SDA integrates processes that extend well beyond the gut, including nutrient absorption, tissue synthesis, ion regulation, and post-absorptive metabolic reorganisation. As a result, it reflects not only digestive work, but the broader allocation of energy toward growth and repair. Across fishes, SDA scales with meal size, temperature, and metabolic phenotype, yet its mechanistic drivers remain debated. Emerging evidence suggests that a substantial proportion of SDA arises from post-absorptive processes, particularly protein turnover and tissue synthesis, rather than the mechanical or secretory costs of digestion alone. This reframing has implications for how we interpret feeding energetics, growth efficiency, and the constraints imposed by environmental stress. In this presentation, I use my research on SDA as a lens through which to revisit the history of this response, interrogate how it is currently interpreted, and explore where a more mechanistic framework may take the field.

DIGESTION The fish gut: Digestion and beyond

Effects of sodium propionate on fatty acid sensing mechanisms in rainbow trout proximal and distal intestinal cell lines

Blanco, Ayelén M. (*amblanco@uvigo.gal*) Poster Presentation
Co-Authors: Fernandez-Maestu, Cristina; Velasco, Cristina; Calo, Jessica and Soengas, José L.
Universidade de Vigo

Abstract: The control of feeding and energy balance in fish relies, among multiple regulatory factors, on the ability of the gastrointestinal tract to detect and integrate luminal nutrient signals. Short-chain fatty acids (SCFAs), such as butyrate, propionate and acetate, are produced by the gut microbiota and are known to play key roles in modulating intestinal nutrient sensing and metabolism in mammals, yet their mechanisms of action in fish remain poorly understood. This study evaluated the effects of sodium propionate on fatty acid sensing in cell lines from rainbow trout (Oncorhynchus mykiss) proximal (RTpiMI) and distal (RTdiMI) intestine. Cell lines were seeded onto 12-well plates, and exposed to different sodium propionate concentrations (250 µM, 500 µM and 1 mM) under serum-fed and serum-deprived conditions for 2, 6 and 24 h. Following treatment, mRNA abundance of key fatty acid transporters and receptors, intracellular signalling components, and enzymes related to lipid metabolism was analysed by qPCR. Propionate exposure modulated the expression of a subset of transcripts associated with membrane transport, receptor-mediated sensing, and downstream metabolic processes. The responses were dependent on both incubation time and concentration, suggesting a dynamic regulatory effect of this SCFA on intestinal cellular pathways. These results provide evidence that

propionate directly influences nutrient-sensing mechanisms in rainbow trout intestinal cells and supports the functional relevance of SCFAs in fish gastrointestinal physiology. Overall, this study contributes to a better understanding of how gut-derived metabolites participate in the integration of peripheral metabolic cues involved in the control of feeding and energy homeostasis in fish.

DIGESTION The fish gut: Digestion and beyond

Warming and food deprivation reshape gut barrier integrity and systemic metabolism in juvenile seabream

Guerreiro, Pedro M[1] (*pmgg@ualg.pt*) Poster Presentation
Co-Author: Tejero, Marc[2]; Gregorio, Silvia F[1]; Fernandez-Alacid, Laura[2] and Ibarz, Antoni[2]
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Abstract: The intestinal epithelium is central to nutrient absorption, ion and water balance, and defense against luminal stressors, making barrier integrity a key determinant of juvenile fish performance. In gilthead seabream (Sparus aurata), elevated temperature and short-term fasting can occur both in aquaculture and in shallow coastal habitats, where juveniles may experience warming events together with reduced food availability. However, their combined effects on intestinal permeability remain poorly understood. We investigated how temperature and feeding status affect intestinal and systemic physiology in juvenile seabream (48 fish; 36.46 ± 0.87 g) maintained at 19, 24, or 29 °C under fed or short-term fasted conditions. Fasting lasted 3–5 days, with shorter deprivation at higher temperature. Intestinal tissues were mounted in Ussing chambers to assess epithelial function through short-circuit current (Isc), resistance, and permeability to FITC-dextran (4 kDa) and RITC-dextran (70 kDa). Plasma osmolality, protein, glucose, lactate, and total lipids were measured together with hepatosomatic index. In parallel, intestinal expression of cldn19, cldn24, and three ocln isoforms was assessed by qPCR. Elevated temperature increased intestinal electrogenic activity, particularly in the anterior intestine, as indicated by higher Isc. Heat also reduced epithelial resistance and increased permeability to both small and large dextran tracers, indicating weaker barrier integrity across intestinal regions. These effects were intensified by fasting. At the whole-animal level, warming and fasting also altered energetic condition. Hepatosomatic index decreased under elevated temperature and food deprivation, indicating reduced energy reserves. Plasma metabolites shifted with treatment: fasting increased total lipids, although this response tended to lessen as temperature rose, while plasma glucose increased at 29 °C, especially in fasted fish. These results show that intestinal permeability and electrophysiological traits are sensitive indicators of gut health under environmental and nutritional stress. Heat intensifies the physiological impact of short-term fasting, promoting increased intestinal activity, barrier dysfunction, and metabolic disturbance.

DIGESTION The fish gut: Digestion and beyond

Intestinal acid-base and nitrogenous waste regulation following blood feeding in sea lamprey, Petromyzon marinus

Quijada-Rodriguez, Alex *aquijadarodriguez@wlu.ca* Poster Presentation
Co-Author: Wilson, Jonathan
Wilfrid Laurier University

Abstract: Sea lamprey are anadromous jawless fish native to the Atlantic coasts of North America and Europe. They are invasive in the Laurentian Great Lakes, where parasitism by landlocked juvenile sea lamprey populations contributed to massive declines in native fisheries. Sea lamprey spend most of their life cycle burrowed in streams as filter-feeding larvae before undergoing a complex metamorphosis into a parasitic blood-feeding juvenile, which migrate downstream to the sea or the Great Lakes where they feed on fish hosts (e.g. salmonids). As parasitic juveniles, sea lamprey can consume as much as 30% of their body mass per day in blood, requiring efficient mechanisms to deal with metabolic wastes (ammonia, urea, and acids) from the host blood and those produced through their catabolism of ingested protein. The goal of this study was to determine how blood-feeding affects acid-base and nitrogenous waste regulatory mechanisms in the intestine of juvenile sea lamprey. We focused on the cellular and molecular mechanisms underlying postprandial regulation of acid-base and nitrogenous waste in the intestine. We found an absence of ammonia transporters along the juvenile sea lamprey intestine. Feeding also caused acidification of the anterior and mid-intestinal regions, coinciding with increased apical/subapical H+-ATPase protein localization in intestinal enterocytes. Overall, the lack of ammonia transporters and increased acidification may function to trap host-derived ammonia in the digestive tract, allowing juvenile sea lamprey to ingest massive amounts of blood during the key juvenile growth phase while limiting uptake of toxic metabolic byproducts in the host's blood.

INVASIVE The physiology, ecology and control of invasive fish species

The physiology, ecology and control of invasive fish species

Mike Wilkie and Pedro Buerreiro

Abstract: After climate change, aquatic invasive species (AIS) pose the biggest threat to biodiversity in both freshwater and marine aquatic ecosystems, and can severely undermine conservation efforts. A key to predicting, preventing and controlling invasive fish species is to develop a better understanding of their physiology. We will assemble a team of internationally recognized experts to discuss our current understanding of the physiology and ecology of invasive fishes focusing on: (i) a review of invasion theory as it relates to the physiology and behaviour of fishes, (ii) physiological features that are common in successful invaders, (iii) use of physiological, ecological and molecular biology approaches to characterize and exploit specific vulnerabilities to mitigate or control invasive species, (iv) the use of eDNA for the early detection and prevention of invasions. Examples of species of interest may include invasive (Asian) carps, goldfish, invasive lake trout and brook trout in western North America, smallmouth and largemouth basses which have been in many novel environments worldwide, sea lamprey and round goby in the Great Lakes, as well as other invasive fishes commonly occurring in Europe pikeperch, bleak, pumpkinseed, the chameleon cichlid or the Wels Catfish. We will also seek out speakers with expertise of invasive species in marine and estuarine habitats including lion fishes and salmonids.

INVASIVE The physiology, ecology and control of invasive fish species

A rainbow trout's guide to recovering from sea lamprey parasitism

Alderman, Sarah[1] (*alderman@uoguelph.ca*) Oral Presentation
Co-Authors: Avey, Sean[1]; Duma, Jessica[1]; Masood, Michelle[1]; Wood, R. Darren[1]; Murphy, Cheryl[2]; Goetz, Rick[3] and Firkus, Tyler[3]
[1]University of Guelph, [2]Michigan State University, [3]University of Wisconsin - Stevens Point

Abstract: In the aftermath of a sea lamprey attack, host fish are faced with loss of body fluids, depleted blood cells, and a gaping wound. Lamprey marks on host fish are used to estimate the abundance of sea lamprey in the Great Lakes, and to inform regulatory decisions aimed at controlling this invasive species. But this external wound divulges little about the sublethal physiological and fitness costs. Using controlled laboratory experiments with sea lamprey and rainbow trout, we quantified changes in a suite of endpoints related to blood composition and function for up to one year after a sublethal sea lamprey attack. Immediately following a 4 day attack, host blood was characterized by a significant drop in plasma protein concentration and depleted leukocytes; however, changes in hematocrit were highly variable, ranging from anemia to polycythemia. Fibrinogen concentration dropped to a third of control levels in parasitized fish which

significantly increased prothrombin (clotting) time and was matched by transcriptional upregulation of fibrinogen alpha and beta chains in the liver. These changes in blood composition and gene expression recovered by 3 months post parasitism, coinciding with complete wound healing. The spleen somatic index (SSI), however, did not fully rebound from the nearly 50% decline immediately after parasitism, as SSI of parasitized fish was also lower than that of non-parasitized fish at 1 year recovery. Combined, these results emphasize the broad impacts of sea lamprey parasitism on host fish physiology, with fitness implications that may persist long after their surface wound has healed.

INVASIVE The physiology, ecology and control of invasive fish species

A Potential Strategy to Offset the Greater Tolerance of Invasive Sea Lamprey to the Lampricide, TFM, in Warmer Waters

Zhao, Zhijun (Tim) (*zhao5090@mylaurier.ca*) Oral Presentation
Co-Authors: Kowalczyk, Sarah; Curkovic, Gabriella; Klepacki, Sebastian; Alsop, Derek and Wilkie, Michael
Wilfrid Laurier University

Abstract: Blood-feeding, invasive sea lamprey (Petromyzon marinus) caused extensive ecological and economic damage to fisheries in the Great Lakes of North America. Populations are now controlled using 3-trifluoromethyl-4-nitrophenol (TFM), a chemical lampricide which selectively targets burrow-dwelling larval sea lamprey in streams. TFM concentrations needed to treat lamprey-infested streams are adjusted based on water pH and alkalinity, which affects TFM bioavailability. Recently, the TFM tolerance of sea lamprey has been shown to increase with temperature, which has not yet been incorporated into treatment protocols. Greater tolerance to TFM at warmer temperatures increases the risk of residual lamprey that survive TFM exposure, and subsequently prey on fishes in the Great Lakes. TFM is often combined with 1% niclosamide during lampricide applications, which increases TFM's toxicity to larval lamprey, but it is not known how this combination is affected by temperature. The goals of this study were to compare temperature-dependent toxicity of TFM to larval lamprey when subjected to treatment TFM or a TFM+1% niclosamide mixtures, and to characterize their associated metabolic disturbances and stress responses. Acute toxicity experiments indicated increased tolerance to TFM at higher temperatures. However, this greater tolerance was attenuated when TFM was mixed with niclosamide. This suggests that niclosamide could be used to mitigate temperature-dependent increases in sea lamprey TFM tolerance, decreasing TFM consumption at warmer temperatures, lessening the risk of residual lampreys, and protecting non-target fishes. Experiments are underway to determine if metabolic disturbances and stress responses due to lampricides are exacerbated at higher temperatures.

INVASIVE The physiology, ecology and control of invasive fish species

Can rock ramp fishways exclude juvenile Common Carp while allowing passage of native fishes?

Myrick, Christopher[1] (*chris.myrick@colostate.edu*) Oral Presentation
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[1]Colorado State University, [2]U.S. Fish and Wildlife Service, [3]West Virginia Department of Natural Resources, [4]Colorado Parks and Wildlife

Abstract: Native fish populations in rivers flowing onto the U.S. Great Plains have been affected by the introduction of nonnative fishes and the presence of dams and irrigation diversions.The latter problem can be mitigated by the installation of fish passage structures that are optimized to maximize passage of the desired native fishes. Unfortunately these structures may facilitate passage of undesired nonnative species such as Common Carp.We measured the effects of slope (4, 6, 8, and 10%) on the passage success of juvenile Common Carp (mean TL 111 ± 12 mm; mean wet weight 17.8 ±6.3 g) in a 6.1-m rock ramp fishway. We then compared the probability of carp passage with that of seven native fishes that occur in systems with the carp to determine if passage structures designed for those species would also facilitate Common Carp passage. Our results show that the probability of juvenile Common Carp passage success in the 6.1-m fishway declines from a high of 0.71 at 4% slope to 0.22 at a 6% slope and drops to zero at both 8 and 10% slopes. However, the Carp had a probability of making it to the 4.06-m mark at 8% and a probability of 0.26 of making it to the 2.03-m mark at 10%. When compared to the native species, juvenile Common Carp have less success at higher slopes (≥ 8%) or in longer fishways (≥ 4 m), raising the possibility of developing fishways that are at least partially selective against the juvenile Carp.

INVASIVE The physiology, ecology and control of invasive fish species

Fine scale response of invasive carp and native fish to an acoustic deterrent at a large riverine lock

Albers, Janice (*jalbers@usgs.gov*) Oral Presentation
Co-Author: Stanton, Jessica; Fritts, Andrea; Hatton, Tyson; Sholtis, Matt; Gibson-Reinemer, Dan; Castro-Santos, Theodore; Woodley, Christa and Brey, Marybeth
U.S. Geological Survey

Abstract: Laboratory studies have suggested that some invasive carps (Hypophthalmichthys spp.) exhibit negative phonotaxis to acoustic signals, in part due to their specialized hearing structure the Weberian apparatus. This behavior has prompted further research into whether these signals could be used at riverine navigation locks to deter upstream movements of invasive carp. In addition, limiting impacts on native fishes from specialized deterrents is an important consideration since navigation locks and dams already pose a major migratory impediment. Underwater acoustic deterrent systems (uADS) installed at lock structures could assist in inhibiting carp movements while limiting

impacts on native fishes, especially if fish respond with predictable behaviors.In 2020, a uADS was installed in the downstream lock approach of Lock No. 19 on the Mississippi River. From 2021 to 2024, the uADS cycled on and off every 80 hours to allow for comparisons of invasive carp and native fish behavior under different operational and environmental conditions. An acoustic telemetry array in the lock approach allowed fine-scale positioning (within meters) of tagged fish. Nearly 900 invasive carp and native fish have been positioned in the study area and used to evaluate fish entrance and exit zones, turn around locations, and locations of deterrent crossings during the on and off deterrent conditions.These results will inform managers on how different fish species interact with the lock structure and the uADS, as well as evaluating the selectivity of the uADS in reducing upstream passage of invasive carps.

INVASIVE The physiology, ecology and control of invasive fish species

Redds, carcasses, and contaminants: ecological impacts of invasive Chinook salmon in South American rivers

Fierro, Pablo[1] (*pablo.fierro@uach.cl*) Oral Presentation
Co-Author: Garcés, Cristobal[1]; Montiel, Sara[1]; Tapia, Jaime[2]; Vega, Carlos[1] and Vargas-Chacoff, Luis[1]
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Abstract: The Chinook salmon (Oncorhynchus tshawytscha) is an invasive species that was intentionally introduced and now maintains self-sustaining populations widely established in rivers of southern South America. During the austral summer and autumn, adults migrate from the ocean to pre-cordilleran rivers, where females construct redds by excavating the riverbed, subsequently dying and contributing marine-derived subsidies. This study evaluated the ecological effects of this reproductive pulse, focusing on benthic habitat disturbance, macroinvertebrate assemblages, organic matter dynamics and the transport of marine-derived metals. In 2024 and 2025, three microhabitats (head, pit, and tailspill) were sampled within redds. Results indicate intense bioturbation, as the excavated pit showed marked reductions in macroinvertebrate richness and density, with dominance of tolerant taxa (Chironomidae and Tubificidae). Organic matter concentration and benthic clorophyll-a also decreased in the pit and tailspill, consistent with sediment removal and resuspension during redds construction. Carcasses are primarily consumed by freshwater crabs, scavenging birds and small mammals, facilitating lateral and longitudinal redistribution of nutrients. Metal concentrations (Cu, Fe, Cr, Mn and Pb) were analyzed in tissues from 30 salmon carcasses (up to three years old, with a maximum length and weight of 102 cm and 11.2 kg). Higher concentrations were found in the liver and in larger individuals. Thus, redds excavation, carcass decomposition, and contaminant release demonstrate that invasive Chinook salmon act as powerful ecosystem engineers in Chilean rivers, altering benthic structure, nutrient dynamics, and metal fluxes across ecosystem boundaries. We thank FONDECYT regular 1240497 and CIN250047 funded by Agencia Nacional de Investigacion y Desarrollo (ANID) Chile.

INVASIVE The physiology, ecology and control of invasive fish species

Born to Survive: Movement Ecology of Invasive Round Goby During Early Life Stages

Zarini, Sina (*s.zarini@outlook.com*) Oral Presentation

Abstract: Early life stages are critical in shaping the dispersal, survival, and population dynamics of aquatic invasive species (AIS). Understanding the physiological and behavioural traits that mediate these outcomes is vital for predicting and controlling successful invaders like the round goby (Neogobius melanostomus). This study investigated how ontogenetic development, diel activity, and environmental cues influence the movement, feeding ecology, and dispersal potential of early juvenile round goby. Laboratory assessments of swimming capacity in small juveniles (<2.5 cm SL) revealed that, despite their small size, these individuals exhibitimpressive locomotor performancethat likely facilitates active dispersal. Furthermore, this research provides the first evidence of diel vertical migration (DVM) of round goby in Lake Ontario. Field surveys and laboratory experiments demonstrated that early juveniles occupy the water surface exclusively at night, a behaviour driven by light cues that likely increases exposure to currents and human-mediated transport vectors. The study further characterized ontogenetic shifts in habitat use, diet, and morphology. Smaller juveniles were more abundant at night in both surface and benthic habitats, consuming primarily pelagic zooplankton. In contrast, larger juveniles transitioned to a primarily benthic existence, feeding on macroinvertebrates. These ecological shifts were mirrored by functional morphological changes, including downward mouth rotation and dorsal eye positioning. Together, these findings provide a mechanistic framework for understanding how the integration of behavioural and physiological traits in early life stages may contributes to the rapid range expansion of AIS, offering potential targets for monitoring and management interventions.

INVASIVE The physiology, ecology and control of invasive fish species

Can biotic resistance to invasions come from behavioural competition? – a case study of gobies from the north east Atlantic

Green, Leon (*leon.green@bioenv.gu.se*) Oral Presentation
University of Gothenburg

Abstract: The spread, impacts and costs from invasive alien species (IAS) are rapidly increasing with growing global shipping and climate change. In the marine environment, management options to combat IAS are virtually absent. Supporting the ecosystems' own resilience, the inherent 'biotic resistance' of the native species community, could become an important tool to help limit IAS. For biotic resistance to occur, niche space in the invaded community needs to encompass the niche space of the invader. Therefore, an invader that shares many traits with species in a community will likely face stronger competition and more difficulty establishing itself there. Here, we share insights into this

phenomenon from several studies on an invasive goby, Neogobius melanostomus, widely distributed in the Great Lakes, the Baltic and estuaries of the NE Atlantic, where we work. We show that despite the invader's reputation as aggressive and territorial, the native species closest in niche-space, Gobius niger, outcompetes the invader in competition trials. Further, we also find the native G. niger to initiate close to four times as many antagonistic interactions against the invasive N. melanostomus, as vice versa. This is likely to have an impact on distributions, which we show with observational data from over 200 video samples from where the species overlap. These two species, their behaviours and relationships shed light on the importance of competition in IAS establishment. Our aim is that these studies will fuel the dialogue on biotic resistance, it's existence and status as a nature based solution tool for invasion management.

INVASIVE The physiology, ecology and control of invasive fish species

Thermal plasticity with physiological trade-offs in the invasive cichlid Australoheros facetus under warming scenarios in Mediterranean-type rivers

Guerreiro, Pedro M.[1] (*pmgg@ualg.pt*) Oral Presentation
Co-Authors: Santos, Emanuel[1]; Costa, Rita A.[1]; Gregorio, Silvia F.[1] and Molina, Juan M.[2]
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Abstract: Climate warming and drought are intensifying thermal stress in Mediterranean freshwater systems, potentially favoring invasive fishes with broad physiological tolerance. We evaluated the thermal biology of the invasive cichlid Australoheros facetus across a wide temperature gradient to test how warming affects metabolic performance, thermal tolerance, and biochemical status. Fish collected from Algarve watercourses were exposed to 5–35 °C, and standard metabolic rate (SMR), maximum metabolic rate (MMR), aerobic scope (AS), Q10, critical thermal maximum (CTmax), and plasma and tissue biomarkers of energy metabolism and oxidative stress were assessed.
Intermediate temperatures (15–25 °C) supported the best overall physiological performance, combining stronger aerobic capacity with higher antioxidant protection. In contrast, 30–35 °C imposed clear physiological costs: maintenance metabolism increased disproportionately, aerobic scope declined, and cellular protection weakened, indicating the onset of heat stress. Despite this, A. facetus showed marked thermal plasticity, with CTmax increasing significantly with acclimation temperature. Fish acclimated to 30 °C had higher CTmax than fish acclimated to 20 °C and 10 °C, although thermal safety margin decreased progressively as acclimation temperature rose. Liver antioxidant activity also peaked at intermediate temperatures and declined at the warmest treatments, reinforcing the mismatch between acute tolerance and sustained performance.
These results show that A. facetus is highly heat tolerant, but that tolerance comes with energetic and cellular trade-offs near upper thermal limits. This combination of broad tolerance and functional performance under warm intermediate conditions may help explain its invasion success and competitive advantage in increasingly hot, low-flow Iberian freshwater ecosystems.

INVASIVE The physiology, ecology and control of invasive fish species

Changes in TFM tolerance & detoxification capacity of the bluegill (Lepomis macrochirus)

Curkovic, Gabriella[1] (*curk1820@mylaurier.ca*) Poster Presentation
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Abstract: Sea lamprey (Petromyzon marinus) invaded the Great Lakes in the late 1800s contributing to severe declines in the populations of native fishes, which were highly vulnerable to these blood-sucking parasites. Since the 1960s, the lampricide 3-trifluoromethyl-4-nitrophenol (TFM) has been added to streams to control larval sea lamprey populations. Recently, larval sea lamprey have been found to be more tolerant to TFM at higher temperatures, suggesting that higher TFM concentrations may be needed to effectively control populations as waters warm seasonally and with climate change. However, this could make non target fishes more vulnerable to TFM toxicity in warmer waters. It is therefore essential to better understand how fishes respond and handle TFM at different temperatures. The bluegill exhibits high TFM tolerance due to its high expression of Phase II detoxification enzymes such as UDP-glucuronylsyltransferases (UDPGTs), which biotransform endogenous and exogenous substances such as TFM to less toxic glucuronidated compounds. However, little is known about how elevated temperatures influence UDPGT enzyme expression, activity, and biotransformation capacity in non-target fishes. To address this gap, acute toxicity tests were conducted at 12°C and 24°C to determine the 12-h LC50 of TFM at each temperature. Results showed that the 12-h LC50 was greater at 24°C, suggesting the bluegill's capacity to detoxify was amplified by temperature. Tissues (muscle, bile, liver) were also collected for TFM quantification and measurement of Phase II enzymes such as UDPGT, which will provide further mechanistic insight into the underlying mechanisms of greater bluegill tolerance to TFM at higher temperatures.

INVASIVE The physiology, ecology and control of invasive fish species

Characterizing the sensitivity of native, non-target lampreys to lampricides used to control invasive sea lamprey (Petromyzon marinus) in the Laurentian Great Lakes.

Kowalczyk, Sarah[1] (*kowa1039@mylaurier.ca*) Poster Presentation
Co-Authors: Padiasek, Lukasz[1]; Borowiec, Brittney[2] and Wilkie, Michael[1]
[1]Wilfrid Laurier University, [2]McMaster University

Abstract: Invasive populations of sea lamprey continue to threaten fisheries and ecosystems in the Laurentian Great Lakes. Populations are controlled by barriers, which block upstream migration of spawning adults, and by chemical control, using the "lampricides" TFM (3-trifluoromethyl-4-nitrophenol) and niclosamide (2',5-dichloro-4'-nitrosalicylanilide). While lampricides are highly effective for sea lamprey control, their

impact on non-target species remains insufficiently characterized. Of particular concern are the four native lamprey species found in the Great Lakes, two of which are of “special concern” due to marked population declines in waterways treated with lampricides. To better characterize the lampricide sensitivity of native lampreys, the acute toxicity and sublethal physiological effects of TFM and a field-relevant TFM-niclosamide (1%) mixture were compared between larval American brook lamprey (Lethenteron appendix) and sea lamprey. We observed that the 12-h LC50 and LC99.9 for both TFM and TFM-niclosamide were indistinguishable between sea lamprey and American brook lamprey, suggesting that this native species is as sensitive to lampricide as sea lamprey. To better understand the mechanisms of lampricide toxicity, we also characterized the physiological effects of sublethal lampricide exposure in different tissues (blood, brain, liver, muscle) to determine if disturbances to energy stores such as glucose, glycogen, ATP, and phosphocreatine were similar to those reported for sea lamprey during and following sublethal lampricide exposure. We conclude that larval American brook lamprey are as sensitive to lampricides as sea lamprey, and that it may be necessary to modify sea lamprey control strategies to minimize adverse effects to these and other native lamprey species.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

The physiology, ecology and control of invasive fish species

Heather Bauer Reid, Vincent Mélançon, Sandra Ann Binning, Mar Pineda and Leon Tran

Abstract: Experimental approaches are key to uncovering complex biological mechanisms. From the lab to the field, experimental studies have facilitated insights into the behaviour, physiology, and ecology of fishes, particularly in response to environmental challenges. Despite their value, findings from experimental studies are seldom translated into conservation practice. This session explores how experimental approaches can be harnessed to inform conservation outcomes. We aim to highlight studies that are designed with conservation in mind, from lab-based approaches that examine individual and group-level responses to environmental change, to applied fieldwork techniques that test interventions in situ. By bridging experimental biology and conservation practices, we hope to foster discussion on how experimental studies can improve ecological predictions and inform decision making processes. We particularly encourage submissions that involve collaborative work with conservation practitioners and broadly welcome studies from all ecosystems.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Oxygen limitation decreases the recovery potential of exploited fish populations; the case of Baltic cod

Behrens, Jane W. (*jabeh@aqua.dtu.dk*) Oral Presentation
Co-Authors: Jacobsen, Nis S.
DTU Aqua

Abstract: Major declines in exploited fish populations are often believed to be driven primarily by overfishing. However, a rapidly changing environment with increasing temperatures and continuously deteriorating oxygen conditions could exert major pressure on fish populations by critically hampering primary productivity drivers: recruitment, growth, maturity and survival. These environmental pressures may even prevent recovery with little to no fishing pressure. Here, we present a case study with Baltic cod using data from a controlled laboratory experiment investigating the effects of prolonged mild hypoxia on growth performance. The study shows that even mild hypoxia resulted in a 30% decrease in average growth. We combine the results from the laboratory study with a population model and show that the estimated decrease in growth will lead to large critical changes in reference points and potential recovery period. The current growth and size at maturity is at an all-time low for Baltic cod, and the targeted fishery has been closed since 2019, limiting data input for stock assessment. The combined effects of not fully enforcing

a fishing moratorium (cod is still frequently caught as bycatch) and deteriorating environmental conditions lead to a poor cocktail for stock recovery. By linking experimental physiology with population-level modeling, this study advances the ecosystem-based approach to fisheries management and highlights the risks of overlooking environmentally-driven productivity changes in rebuilding strategies

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MECHANISM Bridging experimental biology and conservation: From mechanism to application

Developmental acclimation to local thermal environments shapes physiological and hormonal warming responses in anemone-associated reef fish

Bauer Reid, Heather[1] (*heatherreid@trentu.ca*) Oral Presentation
Co-Author: Beldade, Ricardo[2]; Raby, Graham[1] and Mills, Suzanne[3]
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Abstract: Tropical reef systems are increasingly threatened by rising ocean temperatures and other anthropogenic stressors. Species with limited mobility, such as anemone- or coral-associated species, are particularly at risk because they remain exposed to the local environmental conditions around their host after settlement. In Moorea, French Polynesia, the threespot damselfish (Dascyllus trimaculatus) and Polynesian anemonefish (Amphiprion maohiensis) are associated with anemones and occupy habitats that differ markedly in temperature, ranging from warm, shallow lagoons to cooler outer reefs from 10-20 m in depth. Our objective was to determine whether developmental acclimation to local thermal conditions enhances physiological resilience to warming,measured through metabolic rates and circulating cortisol. Juveniles of each species from both habitat types (lagoon and outer reef) were assessed for respirometry as well as baseline and stress-induced cortisol soon after capture under ambient temperature conditions (28 ˚C). Individuals were then split into two groups and either held at 28 ˚C or acclimated to 32 ˚C, a predicted end of century temperature which can induce anemone bleaching and is currently considered abnormally high. Respirometry was then retested after approximately 10 days, as were cortisol levels. By comparing baseline and thermally-induced responses across origin and temperature treatments, the results of this study will elucidate the responses of two vulnerable fish species to future warming. With limited dispersal capability past the larval phase, these findings provide insight into the adaptive potential of site-attached reef fishes via developmental acclimation and highlight that resilience to warming may vary substantially across reef zones, even across small spatial scales.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Projecting habitat suitability for the convict tang (Acanthurus triostegus) in Hawai'i under future ocean conditions using physiology-based models

Tran, Leon[1] (*leontran@hawaii.edu*) Oral Presentation
Co-Authors: Franklin, Erik[1]; Kindinger, Tye[2]; McManus, Lisa[1] and Johansen, Jacob[1]
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Abstract: As ocean warming alters the physical environment of coral reef ecosystems, habitat quality for coral reef fishes grows increasingly uncertain. The physiology and habitat usage of these ectothermic species directly depends on water temperature and will likely be impacted by projected ocean warming. Numerical tools like habitat suitability models are used to determine areas with favorable conditions for species persistence on regional scales by correlating species occurrence with environmental conditions in specific locales. However, the correlative approach of current model frameworks yields high uncertainty when extrapolating to novel ocean conditions. Here, we introduce a multidisciplinary approach to reduce this uncertainty by incorporating physiological processes underlying organismal response to environmental change. We use experimental data on physiological traits in response to warming as a predictor in a suite of habitat suitability models for the convict tang (Acanthurus triostegus). In this work, we 1) parameterize aerobic metabolism with present and projected temperatures; 2) construct habitat suitability models using metabolic responses, environmental conditions, and oceanographic processes to project thermal habitat suitability through the end of the century in the main Hawaiian Islands under SSP1-2.6, SSP2-4.5, and SSP3-7.0. This work aims to inform ecosystem-based fisheries management by planning for future uncertainties in habitat suitability for coral reef fish as ocean warming progresses.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Seasonal thermal tolerance, growth and acclimation capacity in newly settled European plaice (Pleuronectes platessa) from Swedish coastal nursery habitats

Tamarit Castro, Elena[1] (*elena.tamarit@gu.se*) Oral Presentation
Co-Authors: Jutfelt, Fredrik[1]; Perry, Diana[2]; Linderholm, Hans[1] and Gullström, Martin[3]
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Abstract: Anthropogenic climate change is increasing mean ocean temperatures and the frequency and intensity of marine heatwaves. Shallow coastal nursery habitats are particularly vulnerable to thermal extremes because limited water depth amplifies temperature fluctuations. Juvenile European plaice (Pleuronectes platessa), a commercially important flatfish, settle in shallow bays in spring and remain until late-summer migration, thereby experiencing peak seasonal temperatures during a critical growth period. We quantified seasonal changes in thermal tolerance by measuring critical

thermal maxima (CTmax) of wild juveniles from spring to late summer, while continuously monitoring in situ temperatures at 1 m depth. We also conducted laboratory acclimation experiments at ecologically relevant temperatures to assess thermal effects on growth rate, and plasticity in thermal tolerance. CTmax increased with seasonal warming, peaking during July and declining again in late summer. Thermal safety margins were approximately 7–8°C in July, when temperatures ≥24°C occurred repeatedly. In laboratory trials, fish held at 24°C for one month exhibited reduced growth compared to fish maintained at 19–21°C, which reflect local mean summer temperatures. The acclimation response ratio was low (~0.2) after 1 month, indicating limited plasticity in upper thermal tolerance. Although juvenile plaice exhibited seasonal increases in thermal tolerance in parallel with ontogenetic development, acclimation capacity appeared limited and temperatures associated with reduced growth were regularly encountered during post-settlement in shallow nursery habitats. As newly settled plaice rely on these shallow habitats to avoid predation, intensifying coastal warming may increasingly compromise growth and nursery habitat function for this species.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Impact of food restriction on the thermal tolerance and recovery rate of a differently acclimated endangered freshwater fish

Levet, Marie[1] (*marie.levet@inrae.fr*) Oral Presentation
Co-Author: Teulier, Loïc[2]; Brigant, Emma[1]; Melou, Mona[2]; Ghinter, Leopold[2] and Sentis, Arnaud[1]
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Abstract: Heatwaves are increasing in frequency and severity, threatening species worldwide. Ectotherms, such as fish, are particularly susceptible to sudden temperature changes as their body temperature depends on their external environment. While thermal acclimation can help mitigate the impact of prolonged exposure to higher temperatures on an organism's metabolism, sudden temperature shifts can impair aerobic performance by increasing metabolic demands. Rising water temperatures can also disturb food webs, leading to reduced prey availability, in turn affecting organisms' food intake and ultimately energy reserves, which are essential to sustain increased energy demands during an acute warming event. However, how food restriction and temperature interact to define an individual's thermal limits remains poorly understood, while being critical for predicting species vulnerability to extreme thermal events. We acclimated critically endangered Rhône streber (Zingel asper) for eight weeks to two temperature regimes (13°C and 23°C) under either ad libitum feeding or a 50% food restriction. To quantify the energetic costs of acute thermal stress, we combined a CTmax protocol with intermittent stop-flow respirometry. Oxygen consumption was measured before, during, and after the CTmax trial to determine metabolic responses and recovery rate following acute warming.These results establish a relationship between food restriction and thermal tolerance, strengthening our

ability to predict how rising river temperatures, by altering food availability, can impact fish resilience to extreme heat events. By improving assessments of thermal vulnerability, our work aims to inform conservation strategies focused on protecting Rhône streber and, more broadly, wild fish populations exposed to critical changes in their living environments.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Microplastics in riverine fishes – from the river to the lab – and back to the river

Shiels, Holly (*holly.shiels@manchester.ac.uk*) Oral Presentation
Co-Authors: Sulaiman, Badiozaman; David, Gift; Woodward, Jamie
University of Manchester

Abstract: Many urban and sub-urban rivers in the UK are highly contaminated with anthropogenic pollutants including microplastics which are thought to negatively impact fish. Although plastics have been widely documented to be present in wild fish, few studies have quantified the impacts on wild fish health. On the other hand, many lab-based studies have looked at the consequences of microplastic pollution for fish health but have used doses that differ markedly from wild population exposure paradigms. Thus, there is a disconnect in our understanding of impacts of microplastics in wild riverine fish. This presentation will explore ways to better couple lab and field studies to tie pollutant presence with effect in wild populations. We investigated the occurrence, assemblage, uptake and bioaccumulation of microplastic in two mid-consumer fish species as well as the bed sediments and macroinvertebrate prey in the River Tame (Upper Mersey catchment), England. Microplastics were identified and quantified using digital microscopy and μ-FTIR spectroscopy. We found that microplastics spread from the environment into the biota with plastic levels higher in the population living near wastewater treatment outflow. We then took the concentrations and polymer types of microplastics assessed from the wild populations and applied them to fish in a controlled lab setting where impacts on behaviour and physiology could be assessed. The presentation will end with a discussion of how these data are being used to pressure environmental regulators and polluting water companies to improve monitoring and water treatment processes to protect the rivers for fish and other users.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Acute shifts in water chemistry during stocking influence nutrient absorption by young-of-year lake sturgeon

Genz, Janet[1] (*jgenz@westga.edu*) Oral Presentation
Co-Authors: Gonzalez, Kelly[2]; Hindman, Trevor[1]; Coleman, Gary[1]; Forkin, Caitlin[1]; Edwards, Chazz and Dichiera, Angelina[2]
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Abstract: Reintroduction of lake sturgeon (Acipenser fulvescens) populations in the Coosa River is dependent on annual stocking of hatchery-reared juveniles. However, ion availability differs between the hatchery and the river, with lower [Ca2+] and higher [Zn+] and [Mg2+] in the natural river water. Environmental water chemistry is an important driver of ionic regulation in larval A. fulvescens and has been shown to impact early growth and development. The differences in water chemistry between the rearing and stocking environments might also influence growth and survival of stocked juveniles. Investigation of these potential effects on the river population is necessary to optimize stocking procedures and maximize stocking success and natural recruitment.
In this study, hatchery-reared juveniles were acutely transferred to river water, mimicking a stocking event. Parameters associated with growth, macronutrient absorption, and epithelial ion transport were measured at 1, 10, and 21 days post-transfer. Growth, condition, and hepatosomatic index did not differ over the exposure duration. However, the food conversion ratio was more efficient in river water at 10 and 21 days post-transfer. Similarly, while gross intestinal anatomy did not change over the 3-week exposure, the proximate composition of digesta 6 hours post-feeding indicated that fish in river water increased glucose absorption. Gene expression results for related transporters further support our conclusions that environmental ion availability influences intestinal function. This information on nutrient absorption and allocation in stocked young-of-year lake sturgeon can improve the effectiveness of stocking programs for the reintroduction and conservation of this species.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Endurance swimming tests to inform entrainment risk

Hannan, Kelly[1] (*kdhannan@ucdavis.edu*) Oral Presentation
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[1]University of California, [2]The U.S. Army Engineer Research and Development Center

Abstract: The San Francisco–San Joaquin watershed is one of the most heavily managed aquatic systems in California, where thousands of water diversions, annual dredging operations, and large-scale pumping facilities have the potential to impact fish populations. These anthropogenic activities can lead to the entrainment of sensitive native

species, including juvenile Green and White Sturgeon (Acipenser medirostris, A. transmontanus), Chinook Salmon (Oncorhynchus tshawytscha), and Delta Smelt (Hypomesus transpacificus). Anadromous juvenile fishes during outmigration are more likely to encounter multiple structures further increasing their likelihood of entrainment. To better understand species and life stage vulnerability to entrainment, we conducted fixed-velocity endurance swimming trials across multiple life stages. From these trials, we developed endurance curves describing time-to-fatigue (min) data across test velocities. We then integrated these data with observed behaviors (e.g., rheotaxis, benthic orientation, tail-propping) to estimate entrainment risk across a range of intake flow velocities. Results indicate that Chinook Salmon of comparable size to sturgeon and Delta Smelt demonstrate stronger swimming abilities overall. Operational guidelines have historically focused on salmonids, however these differences highlight the importance of species- and size-specific thresholds when developing operational guidance for flow management. By linking laboratory-derived swimming performance data with hydraulic conditions in the field, this work provides a quantitative framework for estimating entrainment risk. These findings inform management decisions regarding flow rates at water diversions, pumping facilities, and dredging operations to minimize entrainment of threatened native fishes and support their conservation.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Physiology, movement, and ecology, oh my! Applying scientific insights to monitor fish at Site C Dam

Pleizier, Naomi (*naomi.pleizier@bchydro.com*) Oral Presentation
Co-Author: Mossop, Brent
BC Hydro, BC, Canada

Abstract: The construction and operation of Site C Dam on the Peace River in northern British Columbia provides an important contemporary opportunity to study the effects of impoundment on inland freshwater fish communities. Monitoring teams have the enormous challenge of quantifying changes in fish abundance and community structure over a large geographic extent as the river transforms into a reservoir. Data collection began multiple years prior to dam completion to characterize baseline abundances of key species. Many monitoring programs will continue for up to 30 years post-construction to document responses of the aquatic ecosystem. I will be discussing the diverse approaches from experimental and observational biology, such as biotelemetry, genetic analysis, eDNA, spawning surveys, and environmental sensing, that are being used to monitor bulltrout movements and spawning abundance, the distribution of kokanee spawning populations, and the effects of total dissolved gas supersaturation on fish health. Though the use of innovative, quantitative monitoring, we hope to document changes in the aquatic community and provide key information for the mitigation, offsetting, and ultimately the stewardship of this valuable ecosystem.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Age is just a number? Exploring the effects of establishment times on fish stress in marine protected areas.

Beach, Riley[1] (*beachr@uwindsor.ca*) Oral Presentation
Co-Author: Radford, Craig[2]; Higgs, Dennis[1]
University of Windsor, Windsor; University of Auckland

Abstract: The Hauraki Gulf Marine Park (HGMP) is a special management area and contains several Marine Protection Areas (MPAs) ranging in age of establishment (from 1975 to 2003). Marine protected areas are designed to safeguard ecosystems from human-driven disturbances and conserve biodiversity. Although reductions in fishing pressure within MPAs have been shown to enhance fish populations and protect species diversity, far less is known about how these protections influence the physiological and molecular stress responses of fish over ecologically meaningful timescales. Traditional physiological and molecular sampling methods are not always permitted for protected species or habitats due to their invasive nature. Repurposing minimally-invasive measures, like swab sampling, can minimize handling, while still collecting meaningful physiological data without the need for invasive tissue samples. Here, we evaluate gill swabs as a minimally-invasive alternative to traditional gill and fin tissue sampling in the Australasian snapper (Chrysophrys auratus). These results suggest that gill swabs can provide a reliable, minimally invasive measure of molecular stress, allowing physiological monitoring of populations in restricted areas, like MPAs within the HGMP. Further, we used gill swabs and fin tissue to evaluate the transcription of stress related genes in three reserves, varying in age of establishment, as well as one non-reserve area. Using a diverse set of locations will allow us to examine transcriptional stress levels across MPAs of different ages.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Bridging physiology and conservation science to promote the recovery of at risk salmonids

Enders, Eva[1] (*eva.enders@inrs.ca*) Oral Presentation
Co-Author: Hnytka, Sarah[2]; Durhack, Travis[2]
[1]INRS, PQ, Canada; [2]Fisheries and Oceans Canada

Abstract: Cold-water salmonids are facing increasing pressure from climate change, habitat alteration, and multiple environmental stressors that threaten their long-term persistence. In response, there is growing interest in integrating physiological data with behavioural insights and habitat models to strengthen conservation planning in rapidly changing ecosystems. Conservation physiology provides a powerful framework for understanding how salmonids respond to environmental variability and future climate conditions. This presentation highlights collaborative case studies on Brook Trout (Salvelinus fontinalis), Athabasca Rainbow Trout (Oncorhynchus mykiss), and Westslope

Cutthroat Trout (Oncorhynchus clarkii lewisi). Each project was co-designed with practitioners to ensure that research questions aligned with conservation priorities and incorporated local ecological knowledge. Our work examined mechanistic, bioenergetic, and molecular responses of these species to changes in temperature, flow, and habitat characteristics. By integrating respirometry and behavioural assays, we linked individual differences in metabolic traits, stress physiology, and movement patterns to environmental quality and climate vulnerability. These datasets generated key parameters for bioenergetic models and species distribution projections, improving our ability to forecast habitat suitability under shifting environmental conditions. Collectively, the research demonstrates how conservation physiology can provide actionable insights for evidence-based management and restoration. The findings support decision making by identifying physiological thresholds that define suitable habitat, informing risk assessments, and guiding recovery strategies under climate-change scenarios. Overall, this work illustrates how integrative physiological approaches can advance both scientific understanding and practical conservation of freshwater salmonids in a rapidly changing world.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Molecular detections of Food Deprivation in juvenile Chinook Salmon: From Lab Development to Field Application

Bugg, William[1,2] (*wbugg@mail.ubc.ca*) Oral Presentation
Co-Author: Greentree, Wes[3]; Innes, Katie[2]; Kahlon, Priyanka[4]; Ming, Tobi[4]; Schulze, Angela[4]; Di Cicco, Emiliano[2]; James, Samantha[2]; Atkinson, Jamieson[5]; Juanes, Francis[3]; Yungwirth, Emily[4]; Curtis, Jennifer[4]; Patterson, David[4]; Bateman, Andrew[2]; Miller, Kristi[4] and Duguid, Will[2]
[1]University of British Columbia, [2]Pacific Salmon Foundation, [3]University of Victoria, [4]Department of Fisheries and Oceans Canada, [5]Coastal Ecosystems Research Collective

Abstract: Along Canada's Pacific coast, cumulative stressors in the early marine environment interact to limit the survival of juvenile Chinook salmon (Oncorhynchus tshawytscha). Prey limitation has been identified across risk assessment frameworks as one of the strongest risk factors for their survival under climate change. In this early marine period, patterns of survival will work to shape population-level recruitment dynamics and future adult returns. However, the early marine period has long been called a 'black box' where environmental and physiological impacts on juveniles are difficult to investigate. To develop molecular tools to investigate food deprivation in the early marine period, we conducted a lab-based food deprivation and refeeding experiment to assess the physiological consequences of starvation. Using samples from this experiment we identified transcriptional responses to develop mRNA-based biomarkers for food limitation that could be assessed using non-lethal gill clips from wild juvenile Chinook salmon. We then used microtrolling to non-lethally capture, sample, and PIT tag juvenile Chinook salmon throughout their early marine migration in the Strait of Georgia (British Columbia) in

the first ocean winter. Using the food deprivation markers, in addition to mRNA signatures of thermal, osmotic, and hypoxia stress as well as pathogens, we assessed the association of these stressors at the time of capture with imminent mortality signatures, and ultimately survival from PIT-tag returns. This presentation will highlight the development of the food deprivation biomarker panel, as well as the findings from its initial application to the non-lethal assessment of stressor signatures in wild juvenile Chinook salmon.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Tracking road salt contamination through community monitoring: Surface water chloride trends in Vancouver streams, and the implications for local salmonids.

Kilgour, Clare L. (*ckilgour@zoology.ubc.ca*) Oral Presentation
Co-Authors: Winter, Carley E.[1]; Hall, Jack[1]; Brauner, Colin J.[1]; Chará-Serna, Ana M.[2]; James, Alan C.[3]; Kroetsch, Nikki C.[4]; Wood, Chris M.[1] and Schulte, Patricia[1]
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Abstract: In northern latitudes, where road salt is applied as a de-icer in winter months, road salt contamination is resulting in the increasing salinization of freshwater streams. We used a network of 29 water quality loggers, across the Vancouver Lower Mainland (VLM), B.C, Canada, to collect high-frequency specific conductance data over five winters. Using specific conductance as a measure of chloride, this data indicates that road salt contamination is causing brief pulses of increased salinity in VLM streams. These road salt pulses can reach concentrations as high as 11x the province's acute guideline for chloride and can occur as frequently as three times per week. Importantly, these pulses occur exclusively between November and March each year, precisely when Pacific salmon species like coho (Oncorhynchus kisutch) and chum (Oncorhynchus keta) are returning to freshwater streams to spawn. As a result, developing Pacific salmon may be exposed to road salt pulses during the earliest and most sensitive periods of their development. In previous laboratory experiments, we have found that the concentrations of road salt regularly seen in streams can be lethal to early life stage Pacific salmon. Therefore, road salt contamination is adding yet another stressor to the gamut for urban Pacific salmon populations. In this analysis, we used laboratory toxicity test data, alongside spawner count information, to better understand the risk being posed to local Pacific salmon populations, with the goal of informing management actions that more effectively protect local salmon-bearing streams (NSERC Alliance).

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Thermal adaptation in freshwater stream fishes

Stanton, Olivia[1] (*oliviastanton02@gmail.com*) Oral Presentation
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Abstract: Temperature is a "master regulator" that determines the physiological performance and behaviour of ectotherms. Thermal tolerance varies widely across species and is thought to be an important factor influencing species distribution. It has been proposed that thermally sensitive species may be at a greater risk of experiencing population declines due to climate change, while invasive species may be particularly thermally tolerant. However, there are few rigorous tests of these ideas. To test these hypotheses, we measured the thermal tolerance of 20 freshwater fish species by measuring the critical thermal maximum (CTmax) and critical thermal minimum (CTmin). We found that both thermal tolerance traits were statistically independent of phylogenetic relationships, indicating phylogeny has little influence on thermal limits. Imperilled species such as the redside dace were not more sensitive than non-imperilled relatives, and invasive species such as the round goby were not more tolerant. Overall, however, we found a significant relationship between CTmax and the southernmost latitude at which a species occurs, suggesting that a high thermal tolerance may allow fishes to inhabit warmer habitats or that fish located in warmer habitats have locally adapted higher thermal limits. These results indicate that thermal tolerance is not the sole predictor of a species' conservation status, but suggest that variation in thermal tolerance is linked to the distribution of ectothermic species. Additionally, these results allow us to better predict which fish species might be sensitive to climate change.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Performance, plasticity, and resilience to warming in diverse populations of brook trout

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Abstract: The effects of climate change are expected to vary across aquatic habitats and geographic areas, so understanding how coldwater fish respond to warming below the species level will be essential for effective conservation and management. Brook trout, thermal generalists among salmonids, exhibit substantial phenotypic plasticity, giving rise to diverse ecotypes and life histories across their wide range, yet this diversity has not been captured in previous work on thermal performance. We investigated how brook trout from 14 populations with distinct ancestries, rearing environments, and life histories responded to thermal stress to elucidate the drivers of intraspecific variation in thermal performance

and identify traits that mediate vulnerability and resilience. To do so, we quantified the metabolic plasticity of over 300 brook trout ranging from 46-448 mm (fork length) by briefly acclimating fish to an optimal or sublethal temperature. Standard and maximum metabolic rates were assessed both at their acclimated temperature and, without further acclimation, at the alternate temperature to evaluate aerobic scope under optimal and thermally challenging conditions. Our measurements of whole-organism performance across diverse populations highlight that thermal tolerance should be considered below the species level, where interactions among ontogeny, life history, and rearing environment jointly shape performance, plasticity, and resilience to environmental change. These findings underscore how experimental approaches that resolve drivers of variation can refine assessments of risk and inform conservation planning.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Acclimation temperature reverses warming-rate effects on upper thermal limits in zebrafish

Ern, Rasmus[1] (*rasmus@ern.dk*) Oral Presentation
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Abstract: We tested how warming rate (1, 5, 9, and 18°C h^{-1}) and acclimation temperature (15, 21, 28, and 35°C) interact to determine acute upper thermal tolerance (critical thermal maximum, CTmax) in zebrafish (Danio rerio). Warming trials were conducted using a custom-built, remotely controlled thermal tolerance assay platform in which fish were housed in individual chambers and monitored via an online video feed, allowing individual recovery protocols to be triggered upon loss of equilibrium through USB-controlled valves and relay switches. Warming-rate effects depended strongly on acclimation temperature: at high acclimation temperatures, CTmax increased with warming rate; at intermediate temperatures, CTmax was largely insensitive to warming rate; and at low acclimation temperatures, CTmax decreased as warming rate increased. These contrasting responses indicate that the physiological processes underlying thermal tolerance adjust during acute warming, and that the rate and magnitude of this within-trial acclimation depend on prior thermal history. Our findings highlight dynamic acclimation as a key determinant of acute thermal tolerance and demonstrate that warming-rate effects cannot be interpreted independently of thermal history, with implications for the comparability of CTmax and thermal death time (TDT) estimates across studies and their use in predictive modelsrelevant to conservation and management decision-making.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Investigating transcriptomic signatures to isolate a cold shock signal from impingement stress in yellow perch

Thapar, Myra[1] (*myrathapar@cmail.carleton.ca*) Oral Presentation
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Abstract: There has been an increasing number of unpredictable cold events that influence lake ecosystems, known as "cold shock", which cause water temperatures to rapidly decline from usual conditions. These events can occur from natural or anthropogenic causes leading to various physiological (e.g. activation of cellular stress pathways) and behavioural (e.g. slowed swimming and impaired escape responses) impacts on fish. Such cold shock events have been seen to coincide with impingement on intake screens of nuclear power plants in the Great Lakes basin, leading to mass mortality of species such as yellow perch (Perca flavescens). Impingement is the entrapment of an organism against intake screens during water intake into power plants. Understanding the relationship between cold stress and impingement is therefore important for both regulators and utilities to better understand causes of fish death and inform changes in mitigation. This study tests whether a molecular signal of cold shock can be isolated from impingement stress. Angled yellow perch were exposed to three different temperatures, a control temperature of 24°C and two cold temperature treatments of 12°C (for 2 or 6 hours), and 6°C (for 2 or 6 hours) followed by simulated impingement for 2 hours. Gill and liver tissues were collected post experimentation. A stress transcriptional profiling chip was used to determine changes in 28 gene profiles post experimentation. Results contribute towards understanding the dynamic relationship between multiple stressors, specifically cold shock and impingement. This work supports and strengthens the knowledge base for decision-making around screening operations and mitigation evaluation.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Minimally invasive sampling for transcriptional profiling to monitor acute stress and recovery in rainbow trout

Jeffries, Ken[1] (*Ken.Jeffries@umanitoba.ca*) Oral Presentation
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Abstract: Transcriptional profiling has significant potential for monitoring wild fishes, which can require non-lethally sampled tissues to reduce harm to fish or to enable post-sampling monitoring. Within captivity, sampling water for transcriptional profiling can provide the physiological status of the fish without needing to physically handle the fish. We have developed a high-throughput qPCR chip that can simultaneously measure 28 genes for 48

individuals in duplicate to monitor stress patterns in mucus for different salmonids. The genes were selected from lab studies that compared multiple tissues to expression profiles of the mucus. However, the time for an acute stress response to be detected in mucus or water is still relatively unknown compared with other tissues typically used for transcriptional profiling. We measured changes in the expression of different stress-related genes over a 4 hr recovery in the mucus and tank water of individual Rainbow Trout (Oncorhynchus mykiss) exposed to a combined acute exercise and thermal stressor. We used the salmonid mucus chip for transcriptional profiling of mucus and water samples. The mucus samples were paired with blood samples that were analyzed for a suite of stress-related indices to compare the recovery between the tissues. The time course of the mucus transcriptional response will inform field sampling of wild fish where there is often a delay between capture and tissue collection. This work provides insight into how long it takes for a stress signature to appear in mucus tissues and in water samples after an acute stressor.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Leveraging empirical science, agent-based modeling, and model sensitivity testing to make informed management decisions that support fish migration in data-poor systems

Dressler, Terra[1] (*tdressler@stillwatersci.com*) Oral Presentation
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Abstract: Establishing safe passage for migratory fish through water management infrastructure is often a priority for resource managers. Mitigation measures to reduce impacts of such infrastructure are often ad-hoc or based on trial and error. Modelling-based approaches to decision making can significantly reduce cost and time to achieve successful mitigation, but managers are often reticent due to concerns over lack of supporting empirical data. Modelling approaches (e.g., agent-based models) that have built-in capacity to evaluate assumptions across a range of uncertainty through sensitivity analyses can allow decision makers to assess the degree to which parameter uncertainty influences the identification of operational best practices. These models can use the best available mechanistic science on fish migration and can identify beneficial future research opportunities. Here, we explore such an approach by combining hydraulic modeling data with existing disparate data on steelhead smolt migration within an agent-based model to evaluate smolt survival and migration success under several operational scenarios for a water diversion in southern California. Model results and sensitivity testing revealed that scenarios providing the best possible outcomes did not change when parameters with high uncertainty were varied across a range of realistic values. This model also revealed unexpected sources of smolt death and identified specific parameters where additional

studies would improve confidence in model outputs. We illustrate that a lack of system-specific data does not preclude using the best available science to establish and evaluate assumptions within decision support models to provide the risk-averse manager with methods to make timely, effective decisions.

MECHANISM Bridging experimental biology and conservation: From mechanism to application

Twenty years on ice: early-life performance following long-term milt cryopreservation in rainbow trout

Kostelnik, Magaleate (*magaleate.kostelnik@wsu.edu*) Oral Presentation
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Abstract: Cryopreservation of fish milt is widely used to preserve genetic diversity and safeguard populations against environmental disturbance and catastrophic loss. However, whether long-term cryopreservation affects offspring performance remains largely untested, limiting our ability to evaluate its conservation utility. If cryopreservation induces sublethal effects, conservation programs may unknowingly produce individuals with compromised physiological performance. This study experimentally assesses whether cryopreservation duration influences early-life fitness in Rainbow Trout (Oncorhynchus mykiss). Eggs from a single maternal pool were fertilized using three treatments: fresh milt (control), 1-day cryopreserved milt, and 20-year cryopreserved milt. Fertilizations were conducted simultaneously, and embryos were reared in replicated incubation units (100 eggs per box; n = 15 per treatment). We quantified fertilization success, survival through yolk absorption, larval growth (body mass and length), and mass-specific standard metabolic rate using intermittent respirometry following yolk absorption. By integrating reproductive success with physiological performance, this study directly evaluates whether long-term cryopreserved milt produces offspring comparable to those derived from fresh gametes. Linking assisted reproductive technologies to mechanistic measures of metabolic function moves beyond fertilization metrics alone and provides critical evidence for assessing the conservation reliability of genetic resource banking. These findings inform the application of cryopreservation in population recovery and conservation breeding programs.