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Jihočeská univerzita
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University of South Bohemia
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Czech Republic

Ph.D. thesis topics 2024/2025
DSP Rybářství / Fishery

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Overcoming barriers in studying gametogenesis using surrogate reproduction

Překonávání bariér ve studiu gametogeneze prostřednictvím náhradních rodičů

Annotation

Surrogate reproduction in fish is achieved via germ stem cell (GSCs) transplantation from a donor into recipient resulting in the production of donor-derived gametes from recipient´s gonads. Nowadays different techniques of GSC transplantation are applied for many different fish species and to facilitate needs of aquaculture biotechnologies as well as nature resources conservation (Lacerda et al., 2006; Yoshizaki et al., 2010).
Proposed PhD topic aims to leverage surrogate reproduction fish to provide access to biological processes in developing gonads which would not be possible otherwise. One of the yet unchallenged frontiers in surrogacy is to use germ cell transplantation as a tool to overcome lethality or augment output of gene editing. There are two prime examples how surrogacy might improve current practices in fish polyploidy induction and analyzing loss of phenotype functions.
First is triploid production using mating tetraploids with diploids. Unfortunately, induced tetraploids are frequently inviable or their overall fitness is heavily impaired. Nevertheless, tetraploidization itself proves feasible, with embryos typically persevering through initial developmental stages. Our preliminary investigations affirm that tetraploid germ cells can be salvaged through transplantation into robust diploid hosts, thereby rendering the method viable for stable triploid production.
Second example is like salvaging tetraploids. Nowadays, gene editing has expanded even into non-model species. However, it remains challenging to perform analysis of loss of function in many genes in particular tissue of interest because they might be essential during embryonic development causing developmental defects or mortality. So far only possible option is to use conditional knock-out employing Cre drivers which are largely unavailable for aquatic models.
This PhD topic aspires to employ gene knockout coupled with subsequent transplantation into hosts to circumvent otherwise inevitable lethality. The targeted genes, such as Sox9 or Nanog, are known for their involvement in gametogenesis, concurrently holding pivotal roles in embryonic development.

PhD student will mainly work in a wet lab and zebrafish facility. The focal points of expertise development include zebrafish culture, *in vitro* reproduction, microinjections, embryology, ploidy manipulation, histology, gene editing, and a spectrum of microscopy techniques.
Aspiring candidates should have background in aquaculture or developmental biology, with hands-on experience in working with aquatic organisms deemed a significant asset. Proficiency in executing routine tasks on embryos utilizing stereomicroscopes is also a desirable attribute.

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The effect of pikeperch (*Sander lucioperca*) origin on their ability to express natural behaviour

Vliv původu candáta obecného (*Sander lucioperca*) na jeho schopnost projevovat přirozené chování

Annotation

Main aim of the study: description of ethological aspects of the pikeperch in controlled conditions with emphasis on the predation, reproductive behaviour, spawning success and influencing factors.

Intensive farming conditions have been shown to significantly impact fish behaviour. The potential impact of rearing history on behaviour may significantly affect the reproductive capacity, survival and predation which may raise concerns about the suitability of fish reared in RAS for release into open water (Ljubobratović et al. 2017). Despite the strong reliance on the expression of natural behaviour in aquaculture, knowledge of the biology of the pikeperch remains limited (Lappalainen et al., 2003; Malinovskyi et al., 2023; Olin et al., 2018). Studies on the reproduction of the (*Sander lucioperca)* is one example, where natural spawning preferences lack description. Another example of behaviour expression in pikeperch is predation, which is one of the main expected outcomes from pikeperch released for top-down control and melioration purposes. Specifically, these fish are anticipated to play a vital role in consuming unwanted small fish, primarily cyprinids, thereby decreasing the risk of zooplankton population reduction and deterioration of water quality. The effect of the origin on the predatory behaviour of pikeperch has not been thoroughly examined to the degree that will allow the assessment of how their origin influences this behaviour. Any identified alterations in behavior may consequently impact their ecological role as predators. Understanding the ethological aspects will help to improve aquaculture culture conditions and promote successfully controlled reproductive cycles (Policar et al. 2019) while answering the question of the suitability of intensive farming to produce pikeperch juvenile fish.

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Increasing operation efficiency of RAS culture of selected high-valuable fish species with the aim to achieve better fish welfare and use system capacity

Zvýšení provozní efektivity chovu RAS u vybraného vysoce hodnotného rybího druhu s cílem dosáhnout lepšího welfare ryb a využití kapacity systému

Annotation

Recirculating Aquaculture Systems (RAS) are rapidly applied around world in the practice of the commercial intensive aquaculture of high-valuable fish species which is not possible effectively cultured in pond aquaculture (Pěnka et al., 2023). RAS aquaculture is technically complicated, demanding on higher investment, production costs, precise technical components, maintenance, and operation. Fish production under RAS is stable in terms of quality and time, predictable around year, but highly depended on water quality of well operating system. Mentioned factors effect profitability and sustainability of RAS aquaculture production in practice (Martins et al., 2010).

The aim of newly opened Ph.D. thesis is:

1) optimize of the selected feeding approach of the selected fish species under RAS;
2) evaluate different therapeutic baths on the performance of biological filtration;
3) testing of different CO_2 concentration in water on efficiency of the selected fish RAS culture;
4) testing of different oxygenation of biological filters for increasing capacity of nitrification;
5) optimize of sedimentation of undissolved substances and their using for natural denitrification unit within RAS.

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Spermatology of endangered, rare ("non-commercial") and invasive fish species of Central Europe and cryopreservation of their sperm

Spermatologie ohrožených, řídce se vyskytujících („nekomerčních“) a invazních druhů ryb střední Evropy a kryokonzervace jejich spermatu

Annotation

Spermatology of commercially important freshwater fish species has received attention for several decades, mainly due to the success of controlled reproduction. Other freshwater fish species have received less attention and for other reasons, such as being readily available as model species. As a result of anthropogenic activity and climate change, changes in the composition of fish communities are occurring, very often in species variation and in the abundance of individuals of particular species. Therefore, varying degrees of protection, support or, on the contrary, reduction of these species come into play, the essential basis of which is knowledge of reproductive biology, not excluding spermatology.

One of the advanced tools for the protection of diversity within a species are so-called gene banks and ex-situ gene pool conservation - which in fish take the form of frozen sperm.

The aim of this work will be to elaborate the spermatological characteristics of selected endangered, rare ("non-commercial") and invasive fish species of Central Europe and to try to develop a procedure for cryopreservation of their sperm for gene bank purposes.

The work requires an active and creative approach, a willingness to learn, knowledge of ichthyology and physiology, and the use of various microscopic and laboratory techniques.

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Circularity and sustainable aquaculture

Cirkularita a udržitelná akvakultura

Annotation

With the rapid growth of the human population, the need for aquaculture fish production increases. Aquaculture has been the fastest-growing sector of animal production in recent decades. This surge in aquaculture raises questions about its sustainability and environmental impact. Most of the concerns about the sustainability of aquaculture are oriented toward feed and the discharge of waste nutrients (water and sludge) into recipients. In the field of feed, the main problems are the lack of traditional feed ingredients – fishmeal and oil, and the so-called "food feed" conflict between the consumption of the same raw materials for human nutrition and feed purposes. In the field of discharge of waste nutrients into the environment, the main problem is the loading of water with nitrogen and phosphorus, which leads to the eutrophication of water. The European strategy FOOD 2030 aims to introduce as many principles of sustainability and circularity as possible into the food production system by 2030. The aim is to reduce the negative impacts of food production on the environment, reduce dependence on fossil fuels, and non-renewable sources of fertilizers, reduce waste production, reduce food feed conflict, and use waste and by-products as much as possible.

As part of the dissertation, the possibilities of using wastewater and sludge from various aquaculture systems will be investigated. Furthermore, the possibilities of using various by-products and waste for fish nutrition will be investigated. Finally, the possibilities of using waste and by-products from fish processing for the production of fish products and feed will be investigated.

Financial support: QK22010177 Optimalisation of supplemental feeding and management of pond aquaculture; 22-18597S Nutrients from fish or nutrition for fish: Unravelling hidden pollution risk and nutrient retention in fishponds by fish nutritional bioenergetic; OP JAK ITI Aquaculture for Future

Feed optimization for sustainable aquaculture

Optimalizace krmiv pro udržitelnou akvakulturu

Annotation

Aquaculture has been the fastest-growing branch of animal food production in recent decades. This surge in aquaculture raises questions about its sustainability and environmental impacts. The feed used usually impacts the farming economy and the environment the most. The main problems are the lack of traditional feed ingredients - fishmeal and oil, the so-called "food feed", the conflict between consuming the same raw materials for human nutrition and feed purposes, and the retention of nutrients in fish gain/loss. Furthermore, for some types of fish, specific stages, or breeding in certain aquaculture systems, there is a lack of information about the optimal diet for its ideal use by the fish organism or other components of the breeding system. Another problem is the occurrence of anti-nutritional substances in alternative feed ingredients, such as trypsin inhibitors, phytates, etc. The aim is to optimize feed in such a way that the nutritional requirements of fish are fulfilled, optimal production and economic

results, balanced high quality of fish, minimal negative impacts on the environment, reducing dependence on insufficient traditional ingredients, reducing "food feed" conflict and using by-products as much as possible.

The possibilities of optimizing feed for different fish species, stages, or breeding systems will be investigated as part of the dissertation. Furthermore, the possibilities of using alternative ingredients or by-products for fish nutrition will be investigated. Last, the possibilities of improving feed use by using probiotics and bioactive substances and reducing the negative effect of anti-nutritional substances will be investigated.

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Fish fins as a welfare indicator

Rybí ploutve jako indikátor welfare

Annotation

In intensive aquaculture systems, the welfare of fish is influenced by many factors as water quality, stocking density, disease management, environmental enrichment, feeding practices, and handling procedures. Maintaining the optimal habitat conditions requires advanced monitoring of additional welfare indicators and stressors manifestations. One of the significant measure of physical health and stressing conditions is the fin morphology.

Damage to fins, whether through nipping, tearing, or fin erosion, can serve as a visible indicator of stress, aggression, or poor environmental conditions. It may compromise the fish's ability to swim efficiently, impacting its navigational skills and making it more vulnerable to predators. Moreover, damaged fins can hinder normal feeding behaviour, reproduction, and social interactions, further affecting the fish's overall health and quality of life. Monitoring and addressing fin damage are critical aspects of ensuring the welfare of captive fish, as it allows for early identification of potential issues, prompt intervention, and the creation of conditions that support optimal fin health and functionality.

Student will collect and annotate diverse dataset of fish fin images, including both healthy and damaged examples, and will be involved in standardizing and augmenting of the dataset. A suitable deep learning model, such as a Convolutional Neural Network (CNN), will be chosen to train and validate the detection and classification. The aim of the students work will be to develop a robust AI model capable of accurately detecting damaged fish fins, contributing to enhanced welfare monitoring in aquaculture systems.

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Molecular background of the developmental switch from planktivory to piscivory in pikeperch brain

Molekulární mechanismy přechodu k dravému způsobu života v mozku mladých candátů

Annotation

The switch from planktivorous foraging to piscivory is one of the most crucial phases in the pikeperch ontogeny since it determines its chances on surviving the first winter. It is a dynamic phase involving already well-known eco-morphologies like shifting pikeperch niche and acceleration of its growth and of the overall development. While ecological aspects of this switch have been investigated for decades the molecular backgrounds remain almost unknown.

There are several possibilities how to utilize already available transcriptomic (RNA-seq) data of two pikeperch generations from the water reservoir Lipno (Czechia). Although the first two RNA-seq batches were performed with the standard poly-A enrichment approach selecting particularly mRNA transcripts, several potentially important non-coding RNA (ncRNA) species have been identified. Hence, further RNA-seq foreseen will be performed via ribodepletion to catch more ncRNAs and active transposable elements beside mRNA transcripts. Moreover, a special attention will be paid to other ncRNA species, including microRNAs, by further modifications of the currently used protocol.

Workflow

1. Field work on the Lipno reservoir – sampling pikeperch brains for RNA isolation
2. Eco-morphological study further exploring the developmental plasticity of young pikeperch
3. Laboratory work – ribodepletion, RNA isolation, sequencing library preparation
4. Bioinformatics – transcriptomic data processing, gene annotation, functional annotation of gene networks, differential gene transcription analysis, alternative splicing analysis, assessment of transcriptional activity of transposable elements, identification of molecular sex marker for juvenile pikeperch utilizing BioProject PRJNA561467 data (de los Ríos-Pérez et al., 2020), integration of all results

Goals

I. Characterization of molecular traits accompanying and following the switch to piscivory
II. Reconstruction of regulatory networks of epigenetic mechanisms governing the gene transcription (i.e. interplay between gene transcription, alternative splicing, and activity of ncRNAs and transposons)
III. Exploration the co-existing distinct size classes in the young-of-the-year pikeperch
III. Establishing machine learning approach in search for patterns in alternative splicing data (optional)

Literature

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