

Topic/Title (Norwegian): Virkningen av forskjellige lysregimer på fiskevelferd

Topic/Title (English): Impact of different light regimes on fish health and welfare

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From left to right: Romain Fontaine, Marco Vindas, Ida Beitnes Johansen, Amin Sayyari

Summary:

Project background: Light is the most stable environmental factor over the years with long days during summer and short days during winter. It has therefore been used by most of animals through evolution to synchronize biological and physiological functions (e.g. growth, reproduction) with the seasons. However, human activities are currently altering this synchronization for many animal species by modifying the light environment.

Indeed, several important Nature articles have recently brought to light the problem for animal life of light pollution in cities. This is a concern for wild as well as farmed animals living near the cities. In fish for instance, it has been shown that ALAN perturb seasonal reproduction along the coast. In addition, in the aquaculture industry, raising fish under continuous light is a common protocol to delay puberty, allowing animals to grow faster. Such environmental conditions have welfare issues for the animals. Indeed, our recent studies show that ALAN and continuous light affect fish behavior, cardiovascular function, sperm quality (two publications in preparations together with the previous master students).

This year, we want to continue to use the medaka, a teleost fish for which we have many tools to further study the effect of different light conditions on its biology and physiology. Interestingly, medaka is a species with a seasonal reproduction. In laboratory conditions (14h of light), this species spawns daily. However, when reducing the light duration to 10h of light, it stops reproducing, making this species interesting to investigate the effect of light on seasonal reproduction. **We thus propose to investigate the effect of different light colors for ALAN/continuous light, and the cellular/molecular mechanisms behind the effects we previously observed on fish heart, spermatogenesis, and behavior.**



Figure 1: Medaka (*Oryzias latipes*)

Hypotheses:

We hypothesize that:

- 1- **different light colors will have different effects on fish biology and welfare**
- 2- **All the changes induced by ALAN or continuous light are triggered by hormonal changes (time and quantity)**

Aim of the project: The main aim of the project is therefore to identify the effect of different light colors and the molecular and cellular changes induced by ALAN and continuous light.

Project plan and implementation: With the previous master student, we went measuring light intensity at different places and depths in Oslo fjord and Akerselva. The student has been interviewed by NRK TV and radio when doing that. In this project, we plan to go back to Oslo to measure light spectrum (color) of the light pollution during the night, also at different places and at different depths. This will provide information on the color of the lights used in big cities in Norway and on how the water absorbs the different colors.

Then, in the model fish facility at the new Veterinary building, we will expose fertilized medaka eggs from different lines (wild type and transgenic lines) to continuous light, to ALAN or to 14h light (control group). 4 colors will be tested (blue, green, yellow, and red).

We will determine the effects of the colors on the parameters we found to show significant differences with the previous student, including stress hormones levels, heart morphology, behavior and sperm quality. We recently obtained a transgenic line of medaka where the blood and heart are fluorescent and thus can be investigated in live animals. We will thus study the effects on the cardiovascular function by counting heart beats and measuring blood flow in live animals using fluorescent microscopy. In addition, using qPCR, we will investigate which molecular and cellular components are involved in the changes induced by ALAN and continuous light. Finally, we will evaluate the sperm quality using computer-assisted sperm analysis (CASA) system.

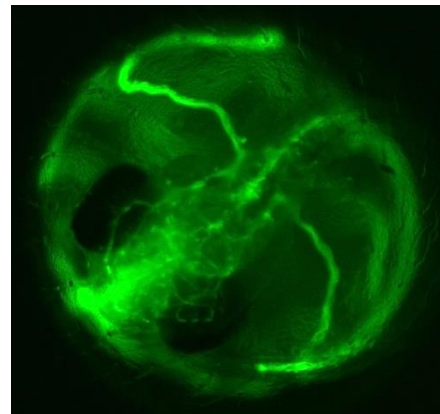


Figure 2: medaka transgenic line with fluorescent heart and blood vessels.

Materials and methods

Animals: A laboratory fish model - medaka (*Oryzias Latipes*).

Medaka (*Oryzias latipes*) is a small, egg-laying, freshwater, bony fish that is native to Asian countries (primarily Japan, Korea, and China). Medaka can easily be maintained and bred under laboratory conditions. At sexual maturity, the body length is about 2.5–3 cm, which under proper rearing conditions is achieved within 2 months after hatching. Spawning is under strict control of light, temperature, and food. In adults, on account of their dimorphic dorsal fins, males are easily distinguished from females.

Methods:

Quantitative Polymerase Chain Reaction (qPCR): The student will learn this regular laboratory technique of molecular biology which monitors the amplification of a targeted DNA molecule during the PCR (i.e., in real time), which provides information on specific gene expression levels in a tissue.

High performance liquid chromatography (HPLC): The student will learn how to use HPLC machine to measure stress related neurohormones (dopamine, serotonin) in brain tissue.

Sperm collection and evaluation using CASA: The student will learn the methods for sperm collection, handling, storage, and evaluation in small model fish.

Blood sampling: The student will learn how to collect blood in a small fish for analysis.



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ELISA: The student will learn how to perform immunoassays (using antibodies) that measure analytes such as the stress hormone cortisol, in a tissue or blood samples.

Microscopy: The student will learn how to use different types of microscopes, including dissection and fluorescent microscopes. This will allow to dissect organs of small size in these small model fish such as the heart and even to count the heart beats in live animals.

Behavioral test: The student will learn how to record fish behavior in different behavioral test setups.

Biostatistics: The student will receive training in how to analyze and run statistics on his data.

Implementation

- The student will be supervised by experts in their field all along the project.
- Dr Ida Beitnes, Dr Marco Vindas, Dr Amin Sayyari, and Dr Romain Fontaine are all localized in the new vet building sharing the same lab and office areas, which facilitates communication, particularly in the coordination and supervision of laboratory techniques.
- All the techniques in this project are already established and regularly used in our lab. The fish are raised in the fish facility located in the basement of the new veterinary building allowing easy access for sampling.
- Our group has ample experience with the supervision of students. We have a very international environment which allows for a varied exciting environment where students flourish and thrive.

Subject area (keywords): model fish, physiology, molecular biology, imaging, anatomy, behavior

Language thesis: English

Bachelor or Master thesis: Master project

Credits: 60 credits

Project/company: not part of another project

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