

## COMPLETED PROJECT CASE STUDY

# OPTIMISING FRESHWATER HEALTH INTERVENTIONS IN SEAWATER SALMON PRODUCTION (OPTIBATH)

### PARTNERS

Nevis Marine, University of Stirling, Loch Duart Ltd, Institute of Marine Research

### PROJECT LEADS

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## BACKGROUND

The Scottish Government's Farmed Fish Health Framework (FFHF) identifies gill health as a major challenge for aquaculture and a significant factor in the survival of marine farmed fish. Improving gill health and safeguarding the health and welfare status of gill-compromised fish are therefore urgent priorities for the Scottish aquaculture sector.

This project focused on improving best practice for the treatment of farmed fish by delivering new knowledge about immersion of fish in ambient freshwater or low-salinity/modified seawater to improve gill health or reduce sealice, alongside assessment of the welfare implications of such treatments, including innovative technologies.

Until recently, the only widely available gill intervention was off-label prescription of hydrogen peroxide, at a reduced dose compared to that authorised for sealice treatment. While treatment with ambient freshwater offers an alternative, securing appropriate water sources, ensuring sufficient volume at remote seawater production facilities, and possessing the physical means to bathe the fish, could be challenging.

In recent years, a number of desalination approaches have been established. Combined with development of suitable fish-handling procedures, this has allowed some producers to introduce low-salinity fish health interventions for the treatment of gill health challenges.

## AIMS AND LIMITATIONS

This project aimed to optimise and validate best practices for the treatment of AGD and sealice using low salinity water. This was to be achieved by improving water quality monitoring and understanding of parameters during bath treatments, using novel modified seawater, improving understanding of freshwater 'lensing' treatment, and through examination of new technologies.

The project started in August 2019; however, the project's third-party technology suppliers were unable to provide their technologies. Therefore, the project was terminated in August 2021 before its goals could be reached, although some learnings were gained.

## Work done

Impacts on fish health and welfare, as measured using a range of novel indicators of fish responses, were to be assessed for low-salinity therapies with an initial salinity target range of 2-7ppt. This was to be achieved through the assessment of standard low-salinity treatments [ambient freshwater (rainwater)+seawater], an innovative bath treatment approach developed by Loch Duart using the Akvafresh modified seawater technology, and also 'lensing' approaches where a layer of low-salinity water is employed to treat fish.

The efficacy of a range of modified approaches to the above treatments was to be investigated, including variations in salinities of the treatment water, variations in water source, variations in duration of bath, and 'conditioning' of water using a novel GIS oxygen membrane technology for oxygenation during treatment, and a GIS CO<sub>2</sub>-stripping technology. Innovative multi-sensor water quality monitoring technology provided by See3 was to be provided in order to allow constant measurement of water quality parameters during treatment (temperature, O<sub>2</sub> saturation, ammonia, pH, CO<sub>2</sub>, and conductivity) to increase understanding of optimal parameters

However, following project initiation, it became clear that the project's third-party technology suppliers, which were intended to act via the project's two commercial partners, would be unable to provide the specified technologies. These technologies were central to realising the project's objectives.

In March 2020, the COVID pandemic brought on-site activities at UK universities to a close. The original academic project lead was furloughed and replaced with a new person going forward. At the same time, salmon farms restricted site access to allow essential personnel only. Moreover, the lead researcher through the subcontracted Norwegian Institute of Marine Research (IMR) also left their post during this period.

Furthermore, Pulcea Ltd became part of AquaPharma and withdrew from the project, together with Nevis Marine Ltd in February 2021.

This set of circumstances prevented the planned sampling and analysis through 2020 into 2021. Hence, by SAIC's request, a revised project plan was submitted in July 2021 taking into account the various changes. Unfortunately, the new plan was not found to sufficiently meet the original objectives, and the project was terminated in August 2021.

As a consequence of all of the above, the sampling and analysis originally planned was severely curtailed, which in turn significantly reduced the conclusions and impact that could be drawn from the limited data collected.

Despite these challenges, the team completed various sampling events. The detailed sampling plan aimed to cover a wide range of indicators, from gill health and sea lice counts to haematological parameters and metabolite analysis, providing a holistic understanding of the effects of low-salinity treatments.

These included one preliminary and three treatment sampling events at Loch Duart's site Badcall between August and November 2019. Key timepoints for sampling included pre-freshwater treatment, immediately post-treatment, 24h after freshwater treatment, and 6-7 days post-treatment. These encompassed biological sampling as well as water quality monitoring parameters such as temperature, O<sub>2</sub> saturation, ammonia, pH, CO<sub>2</sub> and conductivity for the purposes of treatment optimisation.

Lethal sampling, involving 80 fish for each treatment, incorporated assessments such as gill scoring, lice count, FISHWELL morphological operational welfare indicators, weight and length measurements, amoebic load determination on gills through swabbing and qPCR, and histology for gill lesion recovery evaluation. Additionally, mucus from the skin surface was sampled for mass spectrometric analysis of metabolites.

Non-lethal sampling, with 160 fish for each treatment, focused on stress evaluation using FISHWELL morphological operational welfare indicators, gill scoring, and lice count for treatment efficacy. Weight and length measurements were also recorded.

Mucus analysis involved non-destructive sampling of skin mucus from 240 individuals, collected in collaboration with IMR, Bergen. Metabolite profiles were determined through mass spectrometric profiling, providing valuable insights into the overall impact of low-salinity treatments on salmon wellbeing in aquaculture settings.

Gill swab and gill tissue qPCR approaches showed high correlation. The former was also significantly correlated with directly observed gill scores. These observations gave confidence that the bath treatments were successfully controlling AGD and reducing impacts on gill health. The low detection/no detection of amoebae seen for the gill tissue samples subjected to the live amoeba qPCR approach may suggest that this gives a better estimate of active amoebae following treatment, as it does not give a signal for nucleic acids from dead amoebae that may lead to lower CT-values.

Amoeba loads were shown to begin recovery by 6-7 days post-treatment (at tested temperatures between 9.2 and 12.6°C), likely indicative of recolonisation of clearing lesions by remaining live amoebae.

Seallice present were *Caligus elongatus*, which are highly mobile as adults with low host specificity and therefore quickly abandoned the host in the low-salinity bath. As they are abundant on over 100 species of marine host and tend to be highly seasonal, they are less likely than *Lepeophtheirus salmonis* to develop tolerance/resistance to low salinity treatments.

Haematological measurements, especially blood cortisol and glucose levels, showed promise in assessing fish welfare during low salinity treatments. However, lactate measurements were less robust, and additional sample data are needed to fully understand stress responses across different treatments.

Most fish exhibited high welfare, but morphological indicators showed minor changes, like scale loss and fin damage, from initial crowding to the treatment's end. No significant deterioration or recovery occurred post-treatment within the 6-7 days of observation. Despite being minor, scale loss could pose risks as entry points for pathogens, and even slight fin damage may offer additional sites for sea louse attachment.

Measurement of treatment exposure times, pH and O<sub>2</sub> saturation were successful. However, the limited number of treatments investigated – three in total – means that no conclusions can be robustly drawn with respect to their possible impacts on fish health and welfare and treatment efficacy, nor concerning clear patterns in the course of a treatment. This situation will be remedied by characterisation of further treatments going forward.

Clear differences seemed apparent in the water chemistry/water quality parameters measured pre- and post-treatment. However, as above, the extremely low sample numbers do not allow for any statistical analysis or for safe conclusions to be drawn.

Mucus sampling and mass spectrometric analysis proved highly productive, showing potential with 21 identified metabolites whose levels significantly changed during the treatment process. These metabolites could serve as markers for fish health/welfare, but further data collection is necessary for a comprehensive examination.

## IMPACT

Despite the circumstances, this project has provided a baseline data concerning low-salinity tarpaulin treatments and demonstrated the potential power of a multi-modal approach in characterising such treatments and their impacts. It has also demonstrated the potential for use of wider approaches, such as the mucus analysis, morphological and gill OWIs and haematological assessments in characterising fish welfare during and following treatments.