



Conference Abstract

# A validated protocol for fish farm monitoring using environmental DNA

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## Abstract

Sea-based fish farms are associated with strong benthic enrichment gradients and routine monitoring is usually required by regulation. Internationally a wide range of approaches exist for measuring the degree of benthic deterioration around fish farming activities, ranging from simple visual or odour assessments to the calculation of secondary indices that combine multiple biological and/or physico-chemical metrics (e.g., AZTI Marine Biotic Index; Invertebrate Species Index; Norwegian Quality Index; Infauna Trophic Index).

In New Zealand, the health of marine benthic ecosystems around coastal salmon farms is currently measured using an Enrichment State (ES) index. This index incorporates physico-chemical (redox, organic matter, sulphates, etc.) and benthic macrofaunal measurements, which requires taxonomic expertise, is time consuming and expensive.

Supported by a range of private/government agencies and industry partners, we have developed and tested the robustness of bacterial, eukaryotic, and multi-trophic Metabarcoding Biotic Indices (b-MBI, e-MBI, and mt-MBI, respectively) using a molecular Eco-Group approach. The indices were calculated via automatic computer pipelines using data collected over a period of nine years from a range of high- and low-flow salmon farms

(12 farms and 60 stations) from three distinct regions in New Zealand. The MBIs were compared against the established ES index.

All MBIs yielded strong and highly significant relationships with the ES index. The strongest relationships ( $R^2 > 0.9$ ) were obtained with the b-MBI. A refinement of the b-MBI (2019-2020) was supported by highly prolific microbes throughout the ES spectrum, and in particular in the upper end of the organic enrichment scale where traditional benthic indices tend to fail. This resulted in ES values of both (molecular-based versus morphology-based) indices to follow a near one-to-one relationship, performing consistently across water flow environments and considered sub-regions. Station-averaged results were also used to compare regulated compliance outcomes between the two indices, based on the current key compliance criteria for farms within each flow regime. Of the 67 seabed monitoring stations that were subsequently classified as compliant or non-compliant, 62 stations had identical compliance outcomes (i.e. 92% of instances). Furthermore, the b-MBI showed consistently narrower (~50%) confidence interval bands when compared to the traditional ES index. The b-MBI offers unprecedented precision for determining subtle changes along enrichment gradients, constituting a valuable asset for triggering timely management responses and improving compliance.

The protocols developed in this project enable rapid, standardised, and cost-effective eDNA isolation and extraction, followed by automatic b-MBI calculation. The affordability and versatility of the b-MBI tool suggests that it could be immediately integrated into current monitoring strategies as the primary benthic assessment tool for assessing benthic impacts of salmon farms in New Zealand.

## **Keywords**

Metabarcoding Biotic Index; Salmon Farms; benthic impacts; Bacteria; Compliance Thresholds; eDNA

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