

## Fish Pathology and Research in Aquaculture **Dr. Madhaviya Gour\***

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### Abstract

One of the main constraints in aquaculture production is farmed fish vulnerability to diseases due to husbandry practices or external factors like pollution, climate changes, or even the alterations in the dynamic of product transactions in this industry. It is though important to better understand and characterize the intervenients in the process of a disease outbreak as these lead to huge economical losses in aquaculture industries. High-throughput technologies like proteomics can be an important characterization tool especially in pathogen identification and the virulence mechanisms related to host-pathogen interactions on disease research and diagnostics that will help to control, prevent, and treat diseases in farmed fish. Proteomics important role is also maximized by its holistic approach to understanding pathogenesis processes and fish responses to external factors like stress or temperature making it one of the most promising tools for fish pathology research [1].

**Keywords:** Proteomics; Fish diseases; Aquaculture; Fish pathology; Fish welfare

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### Introduction

The demand for animal protein for human consumption is rising as a result of an exponential increase in the world population. Aquaculture is becoming an increasingly important source of protein available for human consumption since is an industry capable of providing solutions to feed a rapidly growing human population and reduce poverty in many countries [2]. To achieve that, the scale of aquaculture production and the range of farmed species have increased dramatically over the last two decades. Live production always comprises a risk for loss due to infectious diseases, with farmed fish, due to husbandry practices in aquaculture, being more vulnerable than wild fish to diseases from a wide range of bacterial, viral, parasitic and fungal infections. Also, the tendency to higher density production systems, the perturbations in ecological systems balance related to pollution and climatic changes, and the expected increase in international transactions of aquaculture products and their derivatives contributed to alterations on the dynamics of interaction between organisms, infectious agents, and people. This influences pathogen rates of replication and proliferation, leading to a broader geographic distribution of pathogenic agents and an increase in species affected by disease outbreaks. This makes disease outbreaks an important constraint to this industry,

with a significant impact on the quality, safety and volume of the fish produced throughout the world, that can lead to market access exclusion and major economic loss or costs to the produce [3].

To address infectious pathologies in farmed fish, approaches like epidemiological studies on main areas of aquatic animal health as transboundary and emerging aquatic animal diseases, animal health surveillance and biosecurity program development should be performed. These are crucial to disease prevalence monitorization, early detection of emerging exotic and new diseases and quality management improvement of aquaculture operations.

Nevertheless, to obtain proper epidemiological models, animal health surveillance and biosecurity programs must integrate environmental information and information from different areas like pathogenesis, disease diagnosis, disease resistance, physiological response to pathogens, pathogen characterization, host immune system responses characterization, disease biomarkers and organism response to disease treatment products.

In aquaculture, inappropriate husbandry conditions, or even standard farming practices, are everyday stressors in culture systems [4]. The allostatic load imposed on the animals can reduce

functioning immune mechanisms, consequently favoring diseases and threatening fish welfare. For instance, drastic changes in water temperature (from 27 °C to either 19–23 °C or 31–35 °C) decreased the immune response and resistance to pathogens in Mozambique tilapia (*Oreochromis mossambicus*). More recently, using a transcriptomics approach, the rearing density in Nile tilapia (*Oreochromis niloticus*) was shown to significantly impact on the susceptibility to the oomycete *Saprolegnia parasitica*. However, the association between husbandry-induced stress and disease is not that straightforward. For example, acute stressors have been reported to enhance or decrease some innate immune responses in fish. On the contrary, chronic stressors have mainly been indicated as immunosuppressors. From a productivity perspective, the health of the fish is often interpreted as “absence of disease”, since from either an ethical or an economic point of view; any disease state is unacceptable for the industry. Therefore, disease prevention and eradication are crucial aspects of a fish farm to ensure the production’s sustainability. Providing optimal welfare conditions, monitoring the health parameters routinely and alleviating stress are necessary steps towards this goal [5].

## Disease Diagnostics

Disease Diagnostics to properly diagnose pathology in aquaculture, we must consider disease as a problem with multiple levels of increasing biological complexity, ranging from environmental to the cell, genome and proteome level.

Disease diagnosis concentric ring, representing layers of disease diagnoses as environment, community, organism, tissue, and omics as a tool to interpret cell/tissue responses (adapted from) [6]. New areas like Proteomics can be an important complement to more classical approaches like pathogen identification, disease symptomatology and histopathological analysis to achieve a good disease diagnosis in aquaculture. In Proteomics, regardless the complexity of the analysed protein mixtures that can range from hundreds, to several thousands of proteins, the major goal is the accurate identification of the highest number of proteins as possible in those mixtures [32]. In gel-based approaches, proteins are first separated by one (1-DE)—or two-dimensional gel electrophoresis (2-DE) and then identified by mass spectrometry, whereas in gel-free approaches (or MS-based) protein mixtures remain in solution prior to protein identification. In each case, protein samples may be digested to peptides by a sequence-specific enzyme, typically trypsin, in a so-called peptide-based “bottom-up” proteomics approach, to distinguish it from the analysis of entire proteins in “top-down” proteomics [7]. Peptide samples can then be separated and analysed by liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS), usually employing electrospray ionization (ESI) as the method to convert the peptides to gas phase ions for MS analysis. Alternatively, peptides samples can be analysed by matrix-assisted laser desorption/ionization (MALDI) time-of-flight (TOF) mass spectrometry. The method of choice will always depend on the main research objective, costs and expertise, with MALDI-TOF MS based strategies being most suited for microbial identification and diagnosis, as a rapid, sensitive and economical in terms of both labour and costs [7]. On the other hand, LC-MS/

MS is most suited for large-scale, systematic characterization of proteomes, e.g., involved in host-pathogen interactions, allowing multiplex sample analysis and quantitation. In the following sections we will discuss in more detail main applications of proteomics in pathogen characterization and in host-pathogen interactions. 3.1. Pathogen Identification Pathogen identification is a key area in disease diagnosis and management. Classical, immunological and molecular methods have been routinely and extensively used to Figure 3. Disease diagnosis concentric ring, representing layers of disease diagnoses as environment, community, organism, tissue, and omics as a tool to interpret cell/tissue responses (adapted from). New areas like Proteomics can be an important complement to more classical approaches like pathogen identification, disease symptomatology and histopathological analysis to achieve a good disease diagnosis in aquaculture. In Proteomics, regardless the complexity of the analysed protein mixtures that can range from hundreds, to several thousands of proteins, the major goal is the accurate identification of the highest number of proteins as possible in those mixtures. In gel-based approaches, proteins are first separated by one (1-DE)—or two-dimensional gel electrophoresis (2-DE) and then identified by mass spectrometry, whereas in gel-free approaches (or MS-based) protein mixtures remain in solution prior to protein identification. In each case, protein samples may be digested to peptides by a sequence-specific enzyme, typically trypsin, in a so-called peptide-based “bottom-up” proteomics approach, to distinguish it from the analysis of entire proteins in “top-down” proteomics [8]. Peptide samples can then be separated and analysed by liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS), usually employing electrospray ionization (ESI) as the method to convert the peptides to gas phase ions for MS analysis. Alternatively, peptide samples can be analysed by matrix-assisted laser desorption/ionization (MALDI) time-of-flight (TOF) mass spectrometry [9]. The method of choice will always depend on the main research objective, costs and expertise, with MALDI-TOF MS based strategies being most suited for microbial identification and diagnosis, as a rapid, sensitive and economical in terms of both labour and costs. On the other hand, LC-MS/MS is most suited for large-scale, systematic characterization of proteomes, e.g., involved in host-pathogen interactions, allowing multiplex sample analysis and quantitation. In the following sections we will discuss in more detail main applications of proteomics in pathogen characterization and in host-pathogen interactions [10].

## Conclusion

Overall, we can look at proteomics as a very promising tool for fish pathology research and diagnostic, allowing a more holistic approach to pathogenesis processes, giving important information on pathogen identification and virulence mechanisms characterization and in host-pathogen interactions, enlightening new stress response routes and previously unknown physiological host responses. However, the use of proteomics in fish aquaculture is still in its early days and limited to some sequenced organisms. Further progress in defining aquacultural proteomes and large-scale datasets from diseased fish and fish pathogens will boost the use of proteomic techniques in

aquaculture, that will lead to new and exciting discoveries on this field. But one of the most promising and interesting areas and one that we believe being the future trend in further understanding

the fish response to pathogens, is the study of the interaction holobiome-host-pathogen, with a strong potential for new and more detailed and integrated knowledge of fish pathogenesis.

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