

pathogen and the disease kinetics, little is known about the molecular mechanisms underlying resistance to this disease.

Within the context of the European H2020 project Vivaldi, an experimental population of Manila clam families was produced, and a batch of juvenile individuals from this population were subjected to a 28-day controlled challenge with *Vibrio tapetis* strain CECT4600. Dual diagnosis was carried out to distinguish between healthy and diseased individuals post-challenge: shells were visually diagnosed for presence of BRD, and a PCR method was adapted to detect *V. tapetis* DNA. Total protein extractions were carried out using whole-body tissue homogenates of healthy and diseased clams and proteins were identified using LC-MS/MS. 2093 protein sequences were matched against a reference transcriptome of the Manila clam, and protein intensities based on label-free quantification were compared to reveal 32 and 55 significant proteins in healthy and diseased clams, respectively. These results provide us with important information regarding the major cell processes and the roles they may play in the resistance of *R. philippinarum* to BRD.

Keywords: *Ruditapes philippinarum*, Brown Ring disease, proteomics, disease resistance, aquaculture

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O-126.

Insights into the microbiota of farmed and wild *Mytilus* SP: Is there a link between bacteria communities and host susceptibility?

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Abstract

Since 2014, *Mytilus* species are affected by mass mortality outbreaks especially in French shellfish farms. A first investigation demonstrated the potential implication of *Vibrio splendidus* through the evidence of a virulent strain 10/068 1T1 able to colonize the blue mussel by bypassing external defense barriers and impairing hemocyte activities.

In this study, we explore the role of microbiota in host susceptibility to microbial disease. Different *Mytilus* species (*M. edulis*, *M. galloprovincialis* and hybrid *M. edulis/M. galloprovincialis*) were sampled from mussel farms impacted by seasonal mortalities and from natural site. Then, we explored 1) the composition of bacterial microbiota, 2) the mussel susceptibility to the pathogen *V. splendidus* 10/068 1T1 and 3) the impact of *Vibrio* infection on microbiota bacteria communities.

Keywords: *Mytilus* sp, microbiota, *V. splendidus*, *Vibrio* infection

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O-127.

Gene encoding enzymes in the urea cycle and polyamine synthesis are modulated during an inflammatory response in rainbow trout (*Oncorhynchus mykiss*)

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Abstract

The urea cycle and the genes encoding the major enzymes is still poorly characterised in teleost fish and to date there is no information as to how these genes are regulated during inflammation. Central to the urea cycle is the metabolism of arginine and its precursor amino acids, ornithine and citrulline. In salmonids, arginine is an essential amino acid as sufficient quantities cannot be synthesised endogenously and must be obtained in the diet. Arginine has roles in both the inflammatory innate immune response and subsequent tissue healing. To further understand the role of the urea cycle and related cycles (polyamine synthesis and nitric oxide production) in teleosts, we characterised gene families encoding the key enzymes in this pathway, their expression during an inflammatory response and changes in the free amino acid levels in the blood plasma following *Aeromonas salmonicida* challenge. Due to two whole genome duplication events in salmonid evolutionary history, several genes in these pathways have paralogous copies, with divergent expression patterns. The modulation of the genes involved in the urea cycle during inflammation could open up new lines of research for both fish health and nutrition.

Keywords: Urea cycle, Polyamines, Arginine, Inflammation, Functional feeds

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O-128.

Effect of different stressors on the expression of glucocorticoid receptor 1 (GR1) and GR2 and their implications in the transcriptional immune response in mucosal surfaces

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Abstract

Cortisol is considered the most common physiological indicator of stress in fish. It plays a role regulating energy homeostasis, osmotic balance maintenance, and metabolic reorganization. As a consequence, cortisol may compromise the normal function of other biological processes including the host defense through a delay or reduction of the immune response. Its mechanism of action is mediated through glucocorticoid receptors (GRs) in responsive cells. In Perciformes like sea bass (*Dicentrarchus labrax*), two receptors, GR1 and GR2 have been described, but in gilthead sea bream (*Sparus aurata*) there is still little information about them. Thus, the aim of this study was to deep into the characterization of GR sequences of seabream and their expression under chronic and acute stress. For this purpose, sea bream were subjected to a high-intensity chronic stress for 40 days and the sequences for GR1 and GR2 were identified by RT-PCR and sequencing. In agreement with previous data in other Perciformes species, a deletion/insertion in the C domain (DNA-binding region) between GR1 and GR2 was found in the nucleotide sequence. Since there are previous antecedents in fish that GR1 and GR2 respond to different cortisol concentration levels, we also aimed to evaluate whether different stressors (in terms of intensity and duration) may differentially modulate their gene expression. To do it, gilthead sea bream were subjected to an acute stressor (1 min air exposure) monitored at 1, 6 and 24 hours post-stress and to a long-term stressor (two crowding stress conditions: 40 and 70 kg/m³) and their response evaluated at 7 and 14 days post-stress. The implications of the stress response upon the GR1 and GR2 expression and their consequences on the expression of immune-related genes was assessed. A differential expression of GR1 and GR2 under the different stress situations was observed, confirming the presence of

two glucocorticoid receptors, and their differential response associated to basal or high cortisol concentrations.

Keywords: Stress, cortisol, Glucocorticoid receptors, mucosal-associated lymphoid tissues, gene expression.

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O-129.

Moritella viscosa in lumpfish (*Cyclopterus lumpus*) and Atlantic salmon (*Salmo salar*)

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Abstract

Winter ulcer disease, caused by *Moritella viscosa*, is a significant problem in cold water salmonid farming, although the bacterium can infect and cause disease in a number of other fish species, such as lumpfish (*Cyclopterus lumpus*). Lumpfish are used as cleaner fish, to eat sea lice from Atlantic salmon (*Salmo salar*) in sea pens. It remains to be established whether *M. viscosa* can be transmitted between the fish species. In this study, we examined whether a salmon isolate of *M. viscosa* could infect and cause disease in lumpfish. We further examined whether a lumpfish isolate of *M. viscosa* could infect and cause disease in salmon. Finally, we examined whether vaccination of salmon with a salmon isolate of *M. viscosa* conferred protection against a lumpfish isolate. The data indicate that while lumpfish appeared to be resistant to a salmon isolate of *M. viscosa*, the salmon could be infected with a lumpfish isolate of *M. viscosa*. Vaccination protected the salmon against the salmon isolate of *M. viscosa* but did not confer sufficient protection to prevent infection with the lumpfish isolate.

Keywords: Atlantic salmon, Lumpfish, *Moritella viscosa*, Vaccine, Experimental infection

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O-130.

IFIT5 participates in the antiviral mechanisms of rainbow trout red blood cells

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Abstract

Recent evidences have demonstrated that rainbow trout nucleated red blood cells (RBCs) halted viral hemorrhagic septicemia rhabdovirus (VHSV) infection inside them. A wide variety of defense mechanisms related to the antiviral immune response have been reported for nucleated

RBCs in response to VHSV exposure. In an attempt to identify the specific rainbow trout RBC proteins that interact directly with VHSV, we characterized the immunoprecipitated (IP) proteome of RBCs exposed to VHSV using an antibody against the N protein of VHSV. The IP proteomic characterization identified 31 proteins by mass spectrometry analysis. Among them, we identified interferon-induced protein with tetratricopeptide repeats 5 (IFIT5), a protein belonging to a family of proteins that are induced after the production of type I interferon, which have recently emerged as important players in antiviral innate immunity response. We confirmed the participation of IFIT5 in the rainbow trout RBC antiviral response by examining the expression profile of IFIT5 in RBCs after VHSV exposure at transcriptional and protein levels. In addition, silencing *ifit5* resulted in a significant increase in VHSV replication in RBCs. Moreover, IFIT5 modulation activity assays performed by modulating the IFIT5 RNA-binding pocket cavity, showed an increase in VHSV replication. In summary, these results suggest a possible role of IFIT5 in the antiviral response of RBCs against VHSV. This work broadens the knowledge of fish nucleated RBCs functions and their role in the immune response to viral infections.

Keywords: Rainbow trout, IFIT5, red blood cells, VHSV, antiviral, immune response.

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O-131.

Analysis of immune gene expression during agd infection/reinfection of Atlantic salmon (*Salmo salar*)

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Abstract

Farmed Atlantic salmon (*Salmo salar*) is one of the most economically important aquaculture species worldwide. Expansion has increased rapidly since the 1980s, and the subsequent industrial scale of production has led to numerous infectious diseases afflicting the marine grow-out stage. Amoebic gill disease (AGD) is a parasitic disease that was first recorded in Tasmania in 1989 and subsequently in Scotland in 2012, and is characterized by raised white lesions on the gills, with high mortality if left untreated. Current treatment methods primarily consist of freshwater or chemical treatment, and it is often necessary to repeat these treatments for the duration of the marine stage due to regular reinfection. To date, most research on analysis of the immune responses elicited has focused on salmon gene expression after first infection, or alternatively after numerous repeated infection/treatment cycles. This study has compared gene expression in gill and head kidney tissue during early infection (up to 15 days) with early reinfection (up to 14 days) post a single reinfection after hydrogen peroxide treatment. Multivariate analysis of the global transcript expression of a panel of ~40 immune genes showed that in the gill tissue, prior to hydrogen peroxide treatment, pro-inflammatory pathways were stimulated, whereas after treatment, anti-microbial peptides were more highly expressed. In the head kidney, differences between pre and post-treatment sampling points were limited, with the most pronounced changes between 15 days post infection, and 1 day post treatment. This model of reinfection allows differences in the expression of an array of genes between pre and post treatment to be elucidated, and improves our knowledge of how the salmon immune system responds to multiple rounds of AGD infection.

Keywords: AGD, reinfection, gene expression, salmon, Scotland

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