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Abstract

Skin is a complex mucosal tissue, which is exposed to the outer environment, interacts with a plethora of environmental insults and represents the first line of defence against many pathogens. Sea lice *Lepeophtheirus salmonis* and *Caligus rogercresseyi* constitute a major threat to farmed salmonids, in both Northern and Southern hemispheres, respectively. We examined the response of skin transcriptome to different levels of infection with *C. rogercresseyi* in Atlantic salmon (*Salmo salar*). The infections were carried out on post- smolts using copepods, with a non- infected, control group. On day 14 post- infection, fish were examined for the total number of lice and grouped into low- and high-infected individuals, with 25 ± 5.3 and 59 ± 9.2 lice per fish, respectively. Skin samples (~100 mg) were collected from the area anterior to the dorsal fin and midway to the lateral line. In total, 36 samples samples –with 12 fish allocated to control, low-infected and high- infected group, accordingly- were subjected to total RNA extraction, followed by library preparation and RNA- seq (Illumina NextSeq 500 platform, single reads 150bp long, sequencing depth 20M reads per sample). Raw reads were aligned to the Atlantic salmon reference genome (GCA_000233375.4 ICSASG_v2) and assessed for differential gene expression using EdgeR. Differentially expressed genes were identified following statistical analysis many of which could be defined as immune response genes. Of interest, genes from encoding mucins were detected as being highly enriched in the infected fish compared to control, suggesting an increase in transcription of this gene family. Other genes of interest included lectins and antimicrobial peptides such as hepcidin and cathelicidin. Many cytokines were also found modulated suggesting a complex immunological response. Full gene set enrichment is being performed to define more comprehensively the immune pathways responding to the lice infection.

Keywords: Atlantic salmon, transcriptomics, *Caligus rogercresseyi*, RNA-Seq, differential gene expression

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O-043.

Pesticide drugs used against sea lice cause transcriptome changes in the early stages of the non-target species *Choromytilus chorus*

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Abstract

Azamethiphos and deltamethrin are pesticide drugs commonly used in Chile as control treatments for sea lice outbreaks in salmon farms. However, their effect on many biological processes of non-target marine species is still unknown. Here we described transcriptome patterns of the mussel *Choromytilus chorus* associated to the effect of these drugs on the early stages of its lifecycle. Trocophore and D larvaes were submitted to continuous exposure to azamethiphos and deltamethrin drugs at 10 and 1000 ppb during 5 days. Samples were collected for RNA-sequencing and transcriptomic analyses were conducted to evaluate gene expression and annotation. Short-read sequencing were conducted in the Ilumina HiSeq platform and long-read sequencing on the Oxford's Nanopore MinION platform. Mortality was not observed in either trocophore or D larvaes exposed to both drugs. Also, fertilization was successful at 80% or more in all treatments. However, thousands of assembled contigs were differentially expressed ($FC > |4|$; $p\text{-value} < 0.01$) in RNA-seq analyses from samples exposed to drugs with respect to control. Time of exposure to drugs triggered larger expression changes with respect to control groups than different chemical concentrations. Annotation by Gene Ontology of differential expressed genes revealed that azamethiphos exposure were mostly associated to changes in genes related to general metabolic processes, while deltamethrin to binding proteins. This study contributes to the understanding of the abnormal effects at molecular level that pesticides over early stages of the mussel *C. chorus* as non-target species and can have potential impacts on the later development of this economically important species.

Keywords: Non-target species, mussel, sea lice, azamethiphos, deltamethrin.

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O-044.

Understanding virulence and pathogenesis in controlled conditions: The case of brown ring disease causing immunodepression in clams

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Abstract

The Brown Ring Disease (BRD) has caused high mortality rates since 1986 in the Manila clam *Venerupis philippinarum*, which was introduced in Western Europe in the 1970s. The causative agent of BRD is a Gram-Negative bacterium, *Vibrio tapetis,* which is also pathogenic to fish. Infection of clams inside the adductor muscle has been done and intra-cellular fate of *Vibrio tapetis* has been investigated inside hemocytes using Transmission Electronic Microscopy. Intracellular multiplication of *V. tapetis* inside hemocytes has been followed during three days, showing that *V. tapetis* could be able to interfere with the autophagy process. Comparative genomic analysis of 17 *Vibrio tapetis* strains allowed identifying a type IV secretion system (T4SS) only present in strains virulent to clams. To further characterize the mechanisms underlying pathogenicity in *V. tapetis* CECT4600, we developed a gene deletion approach to determine the role of different genes in infection. In particular, we created a Δ *virB4* null mutant by deleting the full *virB4* gene, which encodes a protein essential to the T4SS assembly and functioning. We performed *in vitro* virulence tests on the wild type and the deleted strains, and showed that the Δ *virB4* mutation cancelled the virulence of *V. tapetis* to clams. These results proved that our approach is appropriate to explore and understand the role of multiple genes in the *V. tapetis* pathogenicity. Our on-going experiments have the objective to further characterize the role of T4SS, as well as other genes, in the *V. tapetis* infection and the development of BRD in the Manila clam.

Keywords: Vibriosis, clams, autophagy, T4SS, mutagenesis.

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