

keywords: Turbot, myeloperoxidase, neutrophils, immune system

Corresponding author.

E-mail address: jesus.lamas@usc.es (J. Lamas).

P-062.

Characterization and function of a group I type I interferon in the cartilage and hard scale fish Chinese sturgeon (*Acipenser sinensis*)

Youshen Li, Panpan Han, Guangming Tian, Lifeng Zhang, Huizhi Guo, Qiaoqing Xu[#].

School of Animal Science, Yangtze University, Jingzhou, China

Abstract

The type I IFNs are a pleiotropic family of cytokines. Three kinds of IFNe were found in the Chinese sturgeon (*Acipenser sinensis*), named with IFNe1, IFNe2 and IFNe3. In the present study, we found that the Chinese sturgeon IFNe2 protein can stimulate the expression of antiviral genes (PKR, VIPERIN, Mx, and ADAR4) and interferon regulatory factors in the Chinese sturgeon fin (CSF) cell, and induce the phosphorylation of IRF3 and IRF7. In addition, IFNe2 can also induce change in self-expression and positively regulate the expression of IFNe3 during early induction. Similarly, IFNe2 can up-regulate the expression of interferon-stimulated genes in EPC cells. EPC cells showed significantly increase antiviral ability when cells were treated with conditioned medium containing Chinese sturgeon IFNe2 for 2 hours prior to SVCV infection. Among the antibacterial activities, we have not found that IFNe2 has a resistance to *Aeromonas hydrophila* isolated from Chinese sturgeon.

Correspondence. School of Animal Science, Yangtze University, Jingzhou, China,

E-mail address: xuqiaoqing@163.com (Q. Xu).

P-063.

Thermal experience during embryogenesis impacts the microRNA transcriptome in the spleen of adult zebrafish (*Danio rerio*)

Q. Zhang[#], I. Babiak, J. Fernandes.

Faculty of Biosciences and Aquaculture, Nord University, 8049 Bodø, Norway

Abstract

The thermal experience during early development is known to have a long-term effect on several adult phenotypes but its impact on the immune system is still poorly understood. MicroRNAs (miRNAs) are a class of small non-coding RNAs that fine-tune various biological processes, including the immune response. Moreover, miRNA expression can be affected by environmental temperature. In this study, we investigated the effect of embryonic incubation temperature (24, 28, or 32 °C for 3, 4 and 5 days, respectively) on the expression of miRNAs in the spleen of adult fish, and their potential involvement in the immune response to lipopolysaccharide (LPS).

Small RNA-seq results revealed that the spleen transcriptome comprised of 150 miRNAs conserved in zebrafish, 130 mature miRNAs known in other species, and 53 novel miRNA candidates. A total of 30 miRNAs were differentially expressed in the spleen of fish from the high (32 °C) embryonic incubation temperature group compared to those from reference temperature group (28 °C). Enrichment analysis showed that the putative target genes of these miRNAs were involved in immune biological processes of “endocytosis”, “vesicle-mediated transport”, “negative regulation of leukocyte activation” and “induction of positive chemotaxis”. No miRNAs were differentially expressed in the low temperature group compared to the reference temperature. LPS challenge induced three miRNAs in the

spleen of fish kept at constant reference temperature. Immune processes such as “endocytosis”, “vesicle-mediated transport”, “cytokine production” and “NIK non-canonical NF-κB signaling” were enriched by their target mRNAs. In conclusion, high embryonic incubation temperature had a long-term effect on miRNA expression in the spleen of adult zebrafish, and the miRNAs differentially expressed with temperature may be involved in fine-tuning immune processes.

keywords: Temperature, miRNA, RNA-seq, immune system, zebrafish

Corresponding author.

E-mail address: qirui.zhang@nord.no (Q. Zhang).

P-064.

Recombinant flagellin B and its ND1 domain from *Vibrio anguillarum* promote in vivo overexpression of IL-1β and IL-8 cytokines in *Salmo salar*

R. González-Stegmaier^{1,2,#}, I. Aguirre³, F. Villarreal-Espíndola^{4,5}, R. Enríquez³, J. Figueroa^{1,2}.

¹ Laboratorio de Biología Molecular de peces. Instituto de Bioquímica y Microbiología. Universidad Austral de Chile, Valdivia, Chile

² Centro FONDAP: Interdisciplinary Center for Aquaculture Research (INCAR), Chile

³ Laboratorio de Biotecnología y Patología Acuática. Instituto de Patología Animal, Universidad Austral de Chile, Valdivia, Chile

⁴ Department of Pathology, School of Medicine, Yale University, New Haven, USA

⁵ Laboratorio Medicina Traslacional. Instituto Clínico Oncológico. Fundación Arturo López Pérez, Santiago, Chile

Abstract

Flagellin is the major component of the flagellum in Gram negative and positive bacteria, it binds and activates the Toll-like receptor 5 and promotes the expression of proinflammatory cytokines and chemokines in vertebrates. As reported, two recombinant molecules of *Vibrio anguillarum*, flagellin (rFLA) and the amino-terminus of the D1 domain (rND1) from the same molecule induce an in vitro upregulation of proinflammatory genes in gilthead seabream and rainbow trout. We have hypothesized that rFLA and rND1 may function as universal immunomodulator molecules in teleost. In this work, we studied in vitro and in vivo the biological properties for each of those molecules in *Salmo salar* and measured proinflammatory cytokines by real time PCR. The results for in vitro assays using SHK-1 cells and isolated head kidney leucocytes (HKL) were comparable and overall showed that IL-8 transcript increased 6-10-fold using rFLA and 2-6-fold using rND1, IL-1β transcript increased 3-4-fold with rFLA and 1.1-1.8 using rND1. We compared the in vivo effectivity of rFLA and rND1 alone or in combination with a commercial vaccine (CV) against *P. salmonis*. IL-1β and IL-8 induction was measured in head kidney at 4, 24, and 72 hours after intraperitoneal (I.P) injection with 5 µg rFLA or 15 µg of rND1. Results showed that rFLA and rND1 induced a time-dependent acute pro-inflammatory response. IL-1β upregulation reached 25-fold above the PBS-control after 4 hours and it decreased progressively until 3 to 6-fold over the baseline. IL-8 showed an acute response, reaching a 13-fold change above basal levels using rFLA or rND1 at 4 hours post IP injection. After 24 hours IL-8 was almost undetectable. The combined challenge (CV plus one single recombinant) showed differential responses based on IL-8 and IL-1β overexpression. For both combinations, an acute IL-8 upregulation of 3-fold change in head kidney after 4 hours was observed. However, the rFLA effect on IL-8 had a shorter duration than rND1 which response was stable until 144 hours after challenge. IL-1β was shortly upregulated by 2-fold by rFLA but not by rND1, and this induction was sustained in time. Altogether, our results suggest that rFLA and rND1 can drive non-redundant cytokines upregulation and both recombinants are valid candidates to be used as an immuno-stimulant or adjuvant in farmed salmon.

FONDECYT POSTDOCTORAL 3170356, FONDAP 15110027, VIDCA UACH.