

P-054.**NKEF, an antiviral protein involved in the immune response of rainbow trout red blood cells**

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Abstract

Nucleated red blood cells (RBCs) of rainbow trout (*Oncorhynchus mykiss*) possess, in addition to the transport of oxygen and carbon dioxide functions, other relevant roles in the immune response. Its participation in the antiviral response against the VHSV (viral hemorrhagic septicaemia virus) is being studied. To establish the proteins of rainbow trout RBCs that interact directly with VHSV, an immunoprecipitate (IP) of the RBCs exposed to VHSV was performed using an antibody against the VHSV N protein. Thirty-one proteins were identified by mass spectrometry analysis and among them the natural killer-enhancing factor (NKEF) was selected. This protein belongs to the family of peroxiredoxins that have an antioxidant function and improve the cytotoxic cells activity. In addition, NKEF has been identified in several species of fish, including the rainbow trout, where its role has been related to oxidative stress and immunity. In order to establish the antiviral role of NKEF in rainbow trout RBCs, the expression profile of NKEF has been studied in RBCs exposed to VHSV. After VHSV-exposure, NKEF was up-regulated at transcriptional and protein levels which correlated with lower levels of VHSV replication. Moreover, the implication of NKEF in the antiviral response of RBCs against VHSV are being evaluated.

keywords: NKEF, peroxiredoxins, immunoprecipitate, VHSV, red blood cells.

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P-055.**The perfect balance: Trade-offs between reproduction and the immune system in reproducing female rainbow trout (*Oncorhynchus mykiss*)**

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Abstract

Optimal allocation of available resources between different physiological functions is fundamental for survival of any organism. Phenotypic plasticity allows, especially in limited resource situations, trade-off between physiological systems. Since both reproduction and immunity are resource-intensive, trade-offs have been studied in different species. Still, there is lack of knowledge on the relation between reproductive and immune systems during the reproductive cycle of a seasonally reproducing fish.

Here we aim to study the reproductive and immune changes during the seasonal reproductive cycle of female rainbow trout (*Oncorhynchus*

mykiss), in order to identify potential trade-offs. To gain insights of the activation of the immune system, fish were immunostimulated with an intraperitoneal injection of LPS.

Our results show a main upregulation of reproductive parameters during the cycle. Immune parameters, in contrary, displayed a general down-regulation during reproduction. Recovery of them 3 months after the spawning moment, with exception of the phagocytic activity, supports the trade-off theory. Moreover, 17 β -estradiol (E2) concentration in plasma was the only parameter significantly correlated with all immune parameters. We had no evidence for an effect of the immunomodulation on the reproductive parameters.

This suggests that reproduction is able to modify immunocompetence, and potentially infection resistance in rainbow trout, and that E2 seems to participate in those immuno-neuro-endocrine interactions.

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P-056.**In vitro study of the serum effect on the supplementation of sea bass (*Dicentrarchus labrax*) head-kidney macrophage culture medium: Effect on the immune response**

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Abstract

Macrophages are the main phagocytic cell in fish and their optimum function is required to protect them against pathogens, playing nutrition an important role in fish resistance to diseases and therefore being an important factor, which affects macrophage function. Aquaculture is a field in continuous growth, which is acquiring high importance due to the increase in the demand of high-quality protein from the rapidly growing world population. Being fish diseases the main cause of economic loss in Aquaculture production, and due to the importance in macrophage activity in the protection against them, fish macrophage function is a key element which has a direct effect on fish welfare and the response of the immune system. In order to evaluate macrophage function, mainly in vivo studies are being performed, while in vitro macrophage culture, with the potential to study and enhance macrophage function, is an attractive field that is yet to be developed and exploited.

The present study was intended to analyze the impacts of the serum in the culture medium enrichment for sea bass head-kidney macrophages, evaluated through the cytokine expression of the macrophages previously stimulated with Poly IC and LPS obtained from *L. anguillarum*. Furthermore, a comparative study between three different diets (Diet 1, fish oil based; diet 2, cameline oil based; diet 3, rapeseed oil based) was performed in order to survey their effect on the immune response expressed by the macrophages cultured in vitro for a period of 72h.

Fish used in this experiment were juvenile sea bass with an average weight of 154g at the moment of kidney extraction, fed with three diets. Both head-kidneys were obtained and head-kidney macrophages were extracted and cultured in vitro both in sea bass serum and fetal bovine serum supplemented L-15 medium. Cytokine expression was measured through qPCR at 0h, 6h, 12h, 24h, 72h post stimulation with Poly IC and LPS. The results obtained through the study showed a stronger and earlier immune response to the Poly IC stimulation in the macrophages cultured in the medium supplemented with Seabass Serum in comparison with the medium supplemented with Fetal Bovine Serum, which is observed both in Diet 1 and Diet 2. Further studies must be performed in order to assess the effect of serum supplementation in macrophage culture medium,