

# Non-Specific Antibodies Induce Lysosomal Activation in Atlantic Salmon Macrophages Infected by *Piscirickettsia salmonis*

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## Abstract

*Piscirickettsia salmonis*, an aggressive intracellular pathogen, is the etiological agent of salmonid rickettsial septicemia (SRS). This is a chronic multisystemic disease that generates high mortalities and large losses in Chilean salmon farming, threatening the sustainability of the salmon industry. Previous reports suggest that *P. salmonis* is able to survive and replicate in salmonid macrophages, inducing an anti-inflammatory environment and a limited lysosomal response that may be associated with host immune evasion mechanisms favoring bacterial survival. Current control and prophylaxis strategies against *P. salmonis* (based on the use of antibiotics and vaccines) have not had the expected success against infection. This makes it urgent to unravel the host-pathogen interaction to develop more effective therapeutic strategies. In this study, we evaluated the effect of treatment with IgM-beads on lysosomal activity in Atlantic salmon macrophage-enriched cell cultures infected with *P. salmonis* by analyzing the lysosomal pH and proteolytic ability through confocal microscopy. The impact of IgM-beads on cytotoxicity induced by *P. salmonis* in infected cells was evaluated by quantification of cell lysis through release of Lactate Dehydrogenase (LDH) activity. Bacterial load was determined by quantification of 16S rDNA copy number by qPCR, and counting of colony-forming units (CFU) present in the extracellular and intracellular environment. Our results suggest that stimulation with antibodies promotes lysosomal activity by lowering lysosomal pH and increasing the proteolytic activity within this organelle. Additionally, incubation with IgM-beads elicits a decrease in bacterial-induced cytotoxicity in infected Atlantic salmon macrophages and reduces the bacterial load. Overall, our results suggest that stimulation of cells infected by *P. salmonis* with IgM-beads reverses the modulation of the lysosomal activity induced by bacterial infection, promoting macrophage survival and bacterial elimination. This work represents a new important evidence to

understand the bacterial evasion mechanisms established by *P. salmonis* and contribute to the development of new effective therapeutic strategies against SRS.

Author keywords

Atlantic salmon

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lysosome activity

macrophages

*P. salmonis*