

Evaluation of gilthead seabream (*Sparus aurata*) immune response after LCDV DNA vaccination

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Introduction: A DNA vaccine against Lymphocystis Disease Virus (LCDV) was developed and its protective efficacy in gilthead seabream (*Sparus aurata*) has been established. The aim of the present study is the evaluation of immune-related gene expression after vaccination to identify which genes could be relevant to control the viral infection.

Methodology: A DNA-vaccine based on the major capsid protein (MCP) of LCDV using pcDNA3.1-NT-GFP-TOPO system was used to carry out the experiment. The vaccine was administered intramuscularly to gilthead seabream specimens (100 g weight) at 10 µg/fish dose. In addition, two control groups, injected with empty TOPO plasmid at the same dose or PBS, were established to evaluate non-specific immune response and basal response of fish, respectively.

In this study 23 genes related to the immune response (*tlr5*, *tlr9*, *ifnI*, *irf1*, *irf3*, *irf9*, *pkr*, *mx1*, *mx2*, *mx3*, *isg15*, *tnfa*, *casp1*, *il1β*, *il6*, *il10*, *ck3*, *ck10*, *c3*, *nccrp1*, *mhcII*, *tcrcβ*, and *ighm*) and 2 reference genes (*ef1α* and *actβ*) were analysed using real-time PCR (RT-qPCR) in samples of head kidney and intestine at 1, 3, and 8 d post-vaccination.

Results: DNA-vaccination of gilthead seabream induced the differential expression of 9 genes in head kidney and 15 genes in intestine samples. Through the course of the experiment, 9 of those genes reached high level of up-regulation comparing to control groups. These genes were related to IFN type I pathway (*irf9* and *mx3*, in head kidney), inflammation (*il1β*, *il6*, *tnfa*, *ck10*, *c3* and *nccrp-1*, in both organs analysed), and adaptive immune response (*mhcII*, in intestine).

Conclusion: The results obtained allow us to understand which genes could be responsible for the protection against LCDV infection conferred by the DNA vaccine in gilthead seabream. Inflammation is the biological process mainly triggered as a systemic response in vaccinated fish. Different gene expression profiles have been observed in each organ, which may indicate specialized roles relative to immune defensive mechanisms.

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