HOW DO NOVEL FEED FORMULATIONS AFFECT GROWTH PERFORMANCE, OXIDATIVE STRESS AND IMMUNE RESPONSE OF ATLANTIC SALMON?

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Introduction

The aquaculture industry continues to grow faster than any other sector of food production. The need to make aquaculture as sustainable and more environmentally conscious as possible is becoming clearer everyday (FAO, 2020). With this in mind, the replacement of fishmeal and fish oil in aquafeeds has been studied in Atlantic salmon (*Salmo salar*) (e.g., Bendiksen et al., 2011) with many products emerging as potential alternatives to conventional ones (e.g., Hodar et al., 2020). One of the main objectives of the EU project GAIN is to evaluate new ingredients that are already commercially available using different formulation concepts that consider all the fish nutritional needs. GAIN diets are based on circular economy principles and maximize resource efficiency, while contributing to zero waste in the agri-food value chain, being cost-effective feeds, and having good social acceptability. The present study aims to understand the actual effects of these novel feed formulations on growth performance, nutritional status, immunity and oxidative status.

Methods

Quadruplicate groups of Atlantic salmon were fed *ad libitum* with three different diets. Two diets were developed to facilitate the eco-intensification of aquaculture through increased circularity and resource utilization (NOPAP - formula without processed animal protein - and PAP - formula with processed animal protein). The third diet was a commercial-like formulation that was used as a control. After a 96-day feeding trial, plasma samples were collected to evaluate humoral parameters (protease, anti-protease, bactericidal activity, and IgM). Liver and head kidney tissues (collected at day 45 and 96) were used for the simultaneous profiling by PCR array of a panel of 38 or 28 genes, respectively, as markers of growth performance, lipid and energy metabolism, and immune and antioxidant activities. Liver samples were also used to analyse lipid peroxidation. In addition, after 45 and 96 days, the lice count and fish welfare were also assessed by standard methods. The dorsal skin and foregut were collected at days 45 and 96 for mucosal mapping (mucous cell area, density, and barrier status).

Results

Growth performance was adequate and comparable to commercial standards for the novel diets tested. Other parameters analysed, including those related to key performance indicators, intestinal and skin dorsal mucosal mapping, plasma innate immune defences, and lipid peroxidation in the liver did not significantly differ across diets. Regarding head kidney gene expression, at Day 45, 2 out of 28 genes in the array were differentially expressed ($p<0.05$). Gene expression of fish fed with novel feed formulations showed a pro-inflammatory profile evidenced by the up regulation of *il-8,* and a down regulation of *il-10*.At Day 96, the same genes continued to be differentially expressed, but gene *clec1b* (membrane protein) was also up-regulated. However, the rest of the analyses do not support this pro-inflammatory profile. A longer trial may bring light to some of the current results. In turn, the liver had a differential gene expression only at the second sampling point (Day 96), where 4 out of 38 genes in the array were affected, including growth performance (*igf2*), lipid metabolism, elongases (*elovl4*), and energy metabolism (*ucp2l* and *sirt1*). These transcriptomic changes may be attributed to an initial response to the experimental diets. Cross-analysis of gene expression by time points and dietary treatment (two-way ANOVA) yielded only 2 out of 38 genes that had significantly different expression across treatments. The differentially expressed genes were related to growth performance (i*gf2*) and lipid metabolism (*elovl4*).

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Conclusions

The novel feed formulations of the GAIN project for Atlantic Salmon seem to be viable options for the near future. In any case, all results are related to the formulation itself and cannot be attributed to a specific ingredient alteration. More studies are necessary to understand the cost-benefit of these new formulations and their market acceptability to optimize sustainability within the current/predictable European regulatory framework.

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