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ABSTRACTS

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ABSTRACTS

DIETARY INCLUSION OF THE MICROALGAE *Phaeodactylum tricornutum* AFFECTS OXIDATIVE STATUS AND GROWTH POTENTIAL IN GILTHEAD SEABREAM *Sparus aurata* JUVENILES

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Introduction

Fishmeal (FM) and fish oil (FO) are the reference protein and fat sources in aquafeeds, but replacing them is industry priority due to sustainability and future availability issues. However, FM and FO are high-quality raw materials and their replacement can lead to impaired fish growth and immune status (e.g., Conceição et al. 2012). FM and FO in addition to balanced amino acid and fatty acid profiles, also bring micronutrients which are often not present in the most used alternative ingredients, e.g. from plant origin. *Phaeodactylum* is a microalgae rich in micronutrients and bioactive compounds such as fucoxanthin and polyunsaturated fatty acids (PUFA) particularly eicosapentaenoic acid (EPA) (Gilbert-López et al., 2014). This study intends to evaluate the effects of dietary supplementation with *Phaeodactylum tricornutum* incorporated in feed by two different processes, either as whole cells (integral cell wall) or broken cells (disrupted cell wall) on fish health status and performance, in a feed formulation scenario of a zero fish meal, reduced fish oil.

Materials and Methods

Three isonitrogenous (46% crude protein) and isolipidic (18% crude fat) extreme diets devoid of FM and reduced FO were formulated. A control (CTRL) diet was supplemented with crystalline amino acids to meet species requirements whereas two other diets comprised the CTRL diet with a 1% inclusion of *Phaeodactylum tricornutum*; diet BC supplemented with microalgae broken cells and diet WC using whole cells. Diets were randomly assigned to triplicate groups of 150 gilthead seabream (*Sparus aurata*) (initial body weight: 13.3 ± 0.3 g) that were fed to satiation twice a day for 92 days. Fish were assigned to 1 m³ tanks. Seawater flow was kept at 2 L.min⁻¹ (mean temperature $23^{\circ}\text{C} \pm 2.6$; mean salinity 34 ± 0.7) in a flow-through system with artificial aeration (mean dissolved oxygen above 5mg.L⁻¹). After 2 and 12 weeks of feeding fish were sacrificed by anaesthesia overdose with 2-phenoxyethanol. Samples of head-kidney and liver were collected and snap frozen in liquid nitrogen. The simultaneous profiling of a panel of thirty-two (in the liver) or twenty-nine (in the head-kidney) genes, considered as biomarkers of fish growth and health performance, were analysed using the Seabream PCR-array platform of Nutrigenomics group of IATS-CSIC (<http://nutrigrp-iats.org>).

Results and Discussion

There were no differences in final body weight between treatments, nonetheless there is tendency for lower growth after 12 weeks in WC group, what may be linked to increasing FCR at 2 weeks in this same group.

IGF-II was significantly up-regulated in WC when compared with the CTRL diet, which might be due to the need of a repairing mechanism, what maybe related to the putative negative effect of WC on growth, especially in the first weeks of feeding. Expression of other genes seems to support this: down-regulation of the molecular chaperon GRP-94; CAPN1 is up-regulated what might indicate an endoplasmic reticulum cellular stress; CAT is significantly up-regulated, which might indicate a reaction to cellular oxidative stress; PGC1 α is up-regulated what reflects increased mitochondrial activity, cellular energy needs/energy demands; UCPI, an uncoupling protein, is significantly up-regulated, which indicates a reduction on mitochondrial efficiency, with a putative role in protection against ROS; and the ratio CAPN1/CAST also indicates cellular stress in fish fed diet WC.

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BC diet does not led to the negative effects of WC, and also seems to increase the availability of nutrients as can shown by a tendency for down-regulation of IGFBP2, a binding protein that regulates the activity of IGFs. This is also supported by down-regulation of IGFBP4, a growth inhibitor. Moreover, biomarkers related with antioxidant effects are significantly down-regulated in BC diet: CAT is significantly down-regulated, probably due to a reduction in oxidative stress; and GPX4, involved in protection of cells against oxidative stress, is also down-regulated.

Conclusions

The present study suggests that the inclusion of *Phaeodactylum tricornutum* whole cells (WC) induces a cellular stress in seabream juveniles, mainly in endoplasmic reticulum (ER). This negative effect seems to be overridden by the process that leads to the broken cells (BC). BC diet not only helps to eliminate the negative effect, but also seems to increase the availability of nutrients.

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