EFFECT OF THE DIETARY INCLUSION OF BLOOD HYDROLYSATES ON EUROPEAN SEABASS RESPONSE TO STRESS

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Introduction

In aquaculture farms, fish are subjected to several stress situations, including high farming densities, periodic handling, routine procedures or even changes in temperature, oxygen and other environmental parameters. All these stressors may induce oxidative stress by causing an imbalance between the generation of reactive oxygen species (ROS) and the scavenging activity of antioxidants [1]. The accumulation of ROS may damage biomolecules such as proteins, lipids or nucleic acid, impairing growth, immune response and flesh quality [2]. Bioactive peptides, small amino acid chains with properties including antioxidant, mineral-binding, immunomodulatory or antimicrobial activities, can be obtained from protein-rich by-products, making them attractive ingredients for inclusion in aquafeeds within the context of a circular economy. Thus, supplementation of diets with functional ingredients able to modulate fish oxidative stress has been considered in this work, using bioactive peptides included in swine blood hydrolysates (BH). We have investigated the potential of the dietary inclusion of BH to improve European seabass (*Dicentrarchus labrax*) oxidative status after air exposure.

Methods

Three fractions of swine BH obtained by autohydrolysis (AH) or enzymatically were selected. The enzymatically obtained BH were further submitted to a micro- (MF) or nanofiltration (NF). Dried hydrolysates were then included in five isolipidic and isoproteic diets for European seabass: a fishmeal (FM) based diet (positive control, PC), a commercial-based diet where 50% of FM was replaced by plant proteins (negative control, NC) and three diets where 3% of each BH was added to the NC. Diets were assigned to triplicate groups of 71 European seabass juveniles (initial weight 12.3 ± 1.4 g), and fed to apparent satiation in a recirculating saltwater system (RAS). After 12 weeks, 9 fish per treatment were either immediately sampled or air-exposed for 1 minute and let to recover for 6 hours prior to sampling. Plasma and liver were collected to evaluate oxidative stress indicators.

Results

Plasmatic cortisol and lactate were elevated for all diets after the air exposure although differences among diets were not found. Glucose levels were unaffected by either diet or stress. Regarding oxidative stress markers in the liver, lipid peroxidation (LPO), measured by TBARS, tended to increase after air exposure. Considering the non-stressed fish, MF

diet led to the lowest LPO levels, being significantly lower than the AH, but without differing significantly from the remaining diets. In stressed fish, the dietary impact on LPO levels was limited. Protein oxidation, evaluated by carbonyl content, decreased in stressed fish, irrespectively of the dietary treatment. Liver catalase was significantly lower in the NC, NF and AH compared to the PC, and increased after stress in all treatments. Glutathione peroxidase was unaffected by diets or stress condition. Fish fed MF had a significantly higher superoxide dismutase activity than those fed PC and AH diets and was reduced in all stressed fish compared to the non-stressed ones.

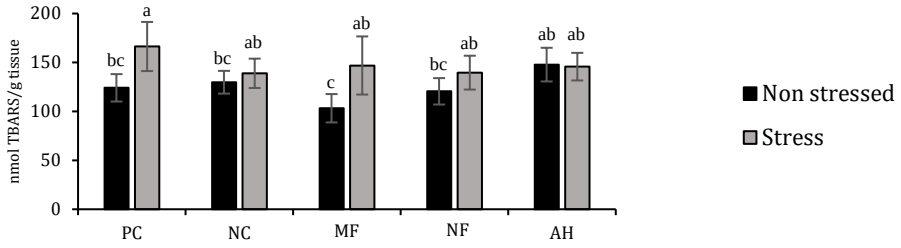

Figure 1 - Lipid peroxidation (LPO) in the liver of fish sampled before (non-stressed) or after (stress) 1 minute air exposure, and after feeding the experimental diets for 12 weeks. Values presented as mean ± standard deviation (n=9). Different lowercase letters indicate significant differences for Diet x Stress after a two-way ANOVA ($p<0.05$).

Conclusions

The stress response triggered by air exposure involved an increase in cortisol levels, which was followed by an increment in plasma lactate; however, the tested BHs could not enhance plasma response to stress. Regarding liver oxidation markers, the stress challenge increased LPO, as ROS can accumulate in the liver under oxidative stress. Carbonyls decreased post-stress, likely due to a feedback interaction with the radicals produced during LPO, which acted to reduce protein oxidation. The NF, composed of smaller peptides than the other BH, may modulate European seabass antioxidant defences by lowering catalase levels without increasing LPO, suggesting this diet could be providing exogenous antioxidants to counteract ROS-induced oxidative stress.

References

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