



The effect of dietary protein replacement by crystalline amino acid on growth and nitrogen utilization of turbot *Scophthalmus maximus* juveniles

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Abstract

A study was conducted to evaluate the effect of partial replacement of dietary fish meal by crystalline amino acids on growth performance, feed utilization, body composition and nitrogen utilization of turbot juveniles.

Four diets were formulated to be isolipidic (12% DM) and isonitrogenous (8% DM). A fish meal based diet was used as control. In the experimental diets, a crystalline amino acid (AA) mixture was used to partially replace fish meal, corresponding to a non-protein nitrogen content of 19, 37 and 56%, respectively (diets 19AA, 37AA and 56AA, respectively). The overall amino acid profile of the experimental diets resembled that of the whole-body protein of turbot. Each experimental diet was fed to triplicate groups of 20 fish (initial body weight of 31.8 g) twice daily to apparent satiation for 42 days. During the trial water temperature averaged 18 °C.

Final body weight, weight gain ($\text{g kg ABW}^{-1} \text{ day}^{-1}$) and specific growth rate were not different between the control and 19AA diet but significantly decreased with the increase of crystalline-AA inclusion from 19 to 56%. Feed intake and feed efficiency of fish fed the control and diet 19AA were similar and significantly higher than those of fish fed the 56AA diet. At the end of the growth trial, there were no significant differences in whole-body composition among groups. Hepatosomatic index was also unaffected by dietary treatments.

Nitrogen retention ($\text{g kg ABW}^{-1} \text{ day}^{-1}$) of fish fed the control and the 19AA diets were similar and significantly higher than that of fish fed the other diets. Expressed as a percentage of the nitrogen intake, N retention was significantly higher with the control than with the 37AA and 56AA diets.

Daily ammonia excretion ($\text{mg kg ABW}^{-1} \text{ day}^{-1}$) of fish fed the control diet was significantly higher than that of fish fed the 37AA and 56AA diets, while daily urea excretion ($\text{mg kg ABW}^{-1} \text{ day}^{-1}$) did not significantly differ among treatments. Non-fecal nitrogen (ammonia+urea) excretion ($\text{mg kg ABW}^{-1} \text{ day}^{-1}$) was significantly higher for fish fed the control diet than in those fed the 37AA and 56AA diets. However, as percent of N intake, ammonia excretion and non-fecal N excretion were significantly higher in fish fed the 56AA diet than in those fed the control and 19AA diets.

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Specific activity of glutamate dehydrogenase, alanine and aspartate aminotransferases did not significantly differ among experimental groups.

In conclusion, in diets with an overall amino acid profile resembling that of the whole-body protein of turbot, crystalline-AA may replace 19% of dietary protein without negatively affecting growth performances or feed utilization efficiency. However, higher protein replacement levels of protein-bound-AA by crystalline-AA severely depressed growth performance.

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1. Introduction

Turbot is a very high market value species and is an important marine fish cultured in Europe. Knowledge of protein utilization by this species is still inadequate, despite its implication on feed cost, protein economy and environmental sustainability. Although, it is reported that turbot has high dietary protein requirement, ranging from more than 50 to 65% of the diet (Cacerez-Martinez et al., 1984; Danielssen and Hjertnes, 1993), there is no available information on essential amino acid (EAA) requirements for this species (Kaushik, 1998). Such information is particularly important to formulate cost-effective diets in which fish meal is replaced by other protein sources.

Although, fish meal is the main protein source in practical diets for turbot, the reduction of dietary fish meal inclusion level is a priority, due to the scarcity of this commodity in the world and consequent rising cost. Comparative to fish meal, alternative protein sources are usually deficient in certain EAA. Therefore, dietary EAA unbalances may occur when these commodities are used in diet formulation (Clarke and Wiseman, 2000). Therefore, the efficient use of alternative protein sources depends on an adequate estimation of EAA requirements of the cultured species.

The reference method to estimate EAA requirement of fish is based on the animal response to increasing levels of dietary incorporation of the EAA under study, being growth or nitrogen retention the response criteria (EIFAC, 1993). In these studies, purified or semi-purified diets are used, in which protein-bound-AAs are replaced by crystalline-AAs. Use of such diets may result in poor growth performances (Cowey, 1995). However, for an unbiased

evaluation of EAA requirement, near optimal fish performance is required. Therefore, it is necessary to confirm the efficacy of crystalline-AAs relative to protein-bound-AAs for fish growth.

The primary objective of the present work was to study the effect of replacing dietary protein-bound-AAs by crystalline-AAs on feed utilization, whole-body composition, nitrogen metabolism and the activity of selected key enzymes of AA metabolism of turbot juveniles. This work will also provide information on the potential use of semi-purified diets for AA requirement studies with turbot.

2. Material and methods

Four experimental diets were formulated to be isolipidic (12% DM) and isonitrogenous (8% DM). A fish meal based diet was used as control. In the other diets, a crystalline-AA mixture was used to replace 25, 50 and 75% of fish meal, corresponding to a non-protein nitrogen content of 19, 37 and 56%, respectively (diets 19AA, 37AA and 56AA, respectively). The overall AA profile, including NEAA, of these diets was made to be equal or higher than that found in the whole-body composition of turbot juveniles as estimated by Kaushik (1998). The crystalline-AA mixture was coated with agar before mixing with the other ingredients. Dietary ingredients were thoroughly mixed and dry pelleted in a laboratory pellet mill (CPM), through a 2 mm die. The pellets were air dried at room temperature and stored in a refrigerator until used. Ingredients and proximate composition of the experimental diets are presented in Table 1. Amino acid composition of the experimental diets is present in Table 2.

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