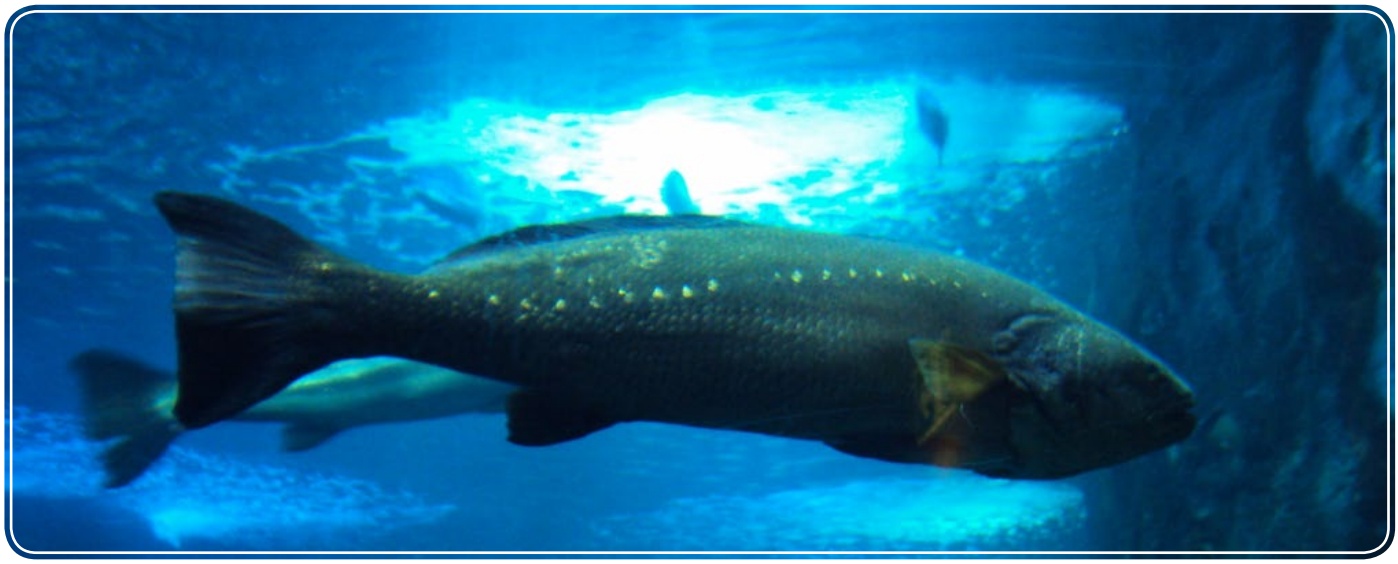
KEY OUTPUT

INVESTIGATING THE BIOSYNTHESIS OF LONG-CHAIN POLYUNSATURATED FATTY ACIDS IN MEAGRE (*ARGYROSOMUS REGIUS*) TO ULTIMATELY ENHANCE THE USE OF VEGETABLE OILS IN MARINE FISH AQUACULTURE



SUMMARY: The biosynthesis of long-chain polyunsaturated fatty acids in meagre (*Argyrosomus regius*) was investigated by characterising both molecularly and functionally the elongation of very long-chain fatty acid-4 (Elovl4) protein. Elovl4 is regarded as a critical enzyme in the biosynthesis of the essential fatty acid docosahexanoic acid (DHA) in marine fish. Meagre were shown to possess both isoforms of Elovl4, namely Elovl4a and Elovl4b.

AT A GLANCE

FULL TITLE: Investigating the biosynthesis of long-chain polyunsaturated fatty acids (LC-PUFAs) in meagre (*Argyrosomus regius*): Functional characterisation of Elovl4 fatty acyl elongases

KNOWLEDGE TYPE: Peer reviewed publication

WHERE TO FIND IT: Not yet available

WHO TO CONTACT FOR MORE INFORMATION: Christiana Kounna, Agricultural University of Athens, Greece, **E:** ccounna@gmail.com, **T:** +306 944 940 776; Dr Oscar Monroig, **E:** oscar.monroig@stir.ac.uk

PATENTS OR OTHER IPR EXPLOITATIONS: No

This research was carried out under AQUAEXCEL Transnational Access in CSIC IATS-ANA, Spain.

INTRODUCTION

Fish, like human beings, require omega-3 (n-3) long-chain polyunsaturated fatty acids (LC-PUFA) for normal growth and development. Fish's natural diet is rich in the n-3 LC-PUFA including eicosapentaenoic (EPA) and docosahexaenoic (DHA) acids, which are found in abundance in fish oils.

In recent times, aquaculture has been subject to sustainability concerns due to its heavy reliance on oils from wild fish as a chief component in feedstuffs. As a result, feed producers have turned to alternative oil sources, such as vegetable oils (VOs).

However, although VOs are rich in C-18 polyunsaturated fatty acids (PUFA), they lack the essential n-3 fatty acids EPA and DHA, which are necessary for marine fish, as these species are regarded as having lower biosynthetic capabilities compared to freshwater and salmonid counterparts. VO-based diets therefore might have a negative effect on fish welfare, as well as on the quality of the end product and therefore for the human

consumer, as long-chain omega-3 contents in the flesh decrease. The biosynthetic ability to convert Carbon-18 to Carbon-20-22 LC-PUFAs varies among species. Carnivorous marine species have low biosynthetic ability for LC-PUFAs. Understanding the biosynthetic pathway of LC-PUFAs will contribute to the understanding of how different dietary products can satisfy the dietary essential fatty acid requirements of marine fish and how the deficiencies of EPA and DHA derived from diets rich in VOs can be met by enhancing the metabolic pathway.

There are a number of genes and enzymes involved in LC-PUFA biosynthesis. This study investigates the molecular basis of the biosynthesis of LC-PUFAs in meagre (*Argyrosomus regius*) through characterisation of genes encoding key enzymes, specifically the elongase of very long-chain fatty acids-4 (Elovl-4). The ultimate aim is to optimise the activity of the biosynthetic pathways and enhance the use of VOs in marine fish aquaculture.

KEY INFORMATION

Meagre has recently been introduced in aquaculture for the purpose of species diversification. Meagre is an interesting species from the perspective of lipid metabolism, as it is a lean fish and previous studies indicate that it accumulates very low levels of mesenteric fat (fat in the fillet, muscle and liver), even when fed on diets with high lipid contents (Chatzifotis *et al.*, 2010; Piccolo *et al.*, 2008), unlike other marine cultured species such as sea bass and sea bream.

The biosynthesis of LC-PUFA in meagre was investigated by characterising both molecularly and functionally the two isoforms of Elovl4 which are found to exist in teleost fish, namely Elovl4a and Elovl4b. Elovl4 is regarded as a critical enzyme in the biosynthesis of the essential fatty acid docosahexanoic acid (DHA), particularly in most marine fish species, which have lost the elongase Elovl2 during evolution.

Due to recent findings that fish possess two isoforms of Elovl4 (a and b), each having a different function, it was decided to introduce this piece of information into this study and characterise both isoforms from meagre. Molecular characterisation was accomplished by molecular cloning of the complementary DNA (cDNA) sequences for Elovl4a and Elovl4b. The protein function was assessed by expressing the open reading frames of both genes in yeast grown in the presence of potential fatty acid substrates.

CONCLUSION

Two distinct genes encoding Elovl4 proteins were isolated in meagre, and were termed as Elovl4a and Elovl4b according to their zebrafish homologues. Both enzymes efficiently elongated PUFAs up to 34 carbons.

END-USER & APPLICATION

- **END-USER 1:** Research organisations related to animal and human nutrition.
  APPLICATIONS: This knowledge can be utilised to gain a better understanding of the molecular mechanisms responsible for the biosynthesis of LC-PUFA in fish. Investigating the ability of endogenous production of these compounds in fish is critical to ensure normal growth and development, as well as welfare of fish in captivity.

- **END-USER 2:** Aquaculture feed industry.
  APPLICATIONS: They can use this information, along with further research, to produce more species specific feeds taking into account the extent to which freshwater and marine fish can utilise alternative sources of lipids to fish oil in diet formulation, without compromising fish health and welfare under culture conditions, and by producing a better quality end product that maintains the nutritional value for the consumers.

- **END-USER 3:** Commercial aquaculture.
  APPLICATIONS: By using feeds that meet the LC-PUFA requirements of meagre, a species that has been recently introduced in aquaculture for species diversification, a good quality end product with sufficient levels of the health-promoting compounds omega-3 fatty acids can be produced.

IMPACT

This study will be of interest to those in the research and scientific community as it is a step towards a more thorough understanding of the LC-PUFA biosynthesis in meagre, and it promotes further research in this area. Further research is also necessary to understand the metabolic pathways of fish more generally. Similar studies could be performed on other species. As a result, animal welfare will be taken into account under culture conditions and good quality products, rich in omega-3, will be produced for consumers. In addition to these results, conducting further research will lead to fully understanding the metabolic pathways involved in LC-PUFA biosynthesis in higher vertebrates.

The knowledge generated in this study has broad application in the aquaculture industry, animal nutrition industry, fisheries, marine biology and environmental management. For example, feed companies can use the knowledge to formulate diets with alternative sources to fish oil which will meet the nutritional requirements of meagre, taking into account the species metabolic requirements known so far from previous studies, the present study and future research. This will improve the culture conditions and the final product, and will alleviate pressure on fisheries for provision of marine raw materials for aquafeeds.