

Corso di Dottorato di Ricerca in Scienze della Vita e dell'Ambiente – Ciclo XL

Biotechnologies applied to aquaculture: functionalization of feed for aquaculture to promote sustainability, profitability and animal welfare.

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Introduction

Aquaculture has emerged as a key pillar in global food security, now accounting for over half of the world's seafood supply. With global demand for fish projected to reach 205 million tons by 2032, the sector continues to expand rapidly. This expansion necessitates the adoption of more efficient, sustainable, and environmentally responsible practices. Within this framework, aquafeeds occupy a pivotal position, as they constitute the largest operational expenditure in intensive aquaculture systems, requiring the refining of feed formulations and delivery strategies. These practices can substantially decrease nutrient waste, improve feed conversion efficiency, and mitigate environmental impacts, including nutrient leaching and the consequences of overfeeding. In fact, ensuring an optimal fish nutrition employing tailored feed formulations represents a key factor in enhancing animal health, growth performance, and product quality. An effective strategy to minimize stress in fish, thereby reducing their susceptibility to infections, can involve the use of bioactive molecules as feed additives designed to enhance fish welfare. However, these substances are often sensitive to environmental stressors such as heat, light, oxygen, and antibiotics. For that reason, encapsulation technology can represent an innovative strategy to preserve the structural and functional integrity of these compounds. This ensures the long-lasting storage and nutrient delivery of a number of compounds, improving the feeding strategies of the aquaculture sector.

Aim & methods

The primary objective of this PhD project is the development and validation of innovative functional aquafeeds aimed at enhancing sustainability, economic efficiency, and fish welfare. To achieve this, advanced biotechnological approaches have been employed, with a particular focus on a novel microencapsulation technology—Co.M.E. (Coating MadeEasy), developed by STM Aquatrade S.r.I.—as the foundation for this work. This technology enables the stable and targeted incorporation of bioactive compounds directly into fish feed, using natural carriers (e.g., starch, gum arabic, maltodextrin) that ensures the preservation. The resulting microcapsules, named +POP, represent a promising solution for the precise and efficient delivery of functional ingredients, potentially improving both the nutraceutical effectiveness of the feed and the physiological response of the target species.

Microencapsulation of selected bioactive compounds in this project:

- **1.** Astaxanthin \rightarrow for flesh pigmentation and antioxidant support for animal welfare;
- **Postbiotics** \rightarrow to promote animal welfare;
- **Rosemary essential oils** -> utilized to develop a new line of naturally flavored trout products, enriched with essential oils and designed to be ready-to-cook.

WP1 - Months 1-2 Production of the Rosmery essential oil at UNICAM

1. Essential oils distillation



2. Chemical profile analysis of the oil via gas chromatography (GC-FID and GC-MS).						
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software from Agilent and the NIST Mass Spectral Search Program



19,8%

monoterpene hydrocarbons

WP2 - Months 3-5 stage at STM Aquatrade

Microencapsulation of molecules into Microcapsules (+POP)

Production of the microcapsules (+POP) with:

Astaxanthin



A six-hour hydrodistillation of 1 kg dry leaves of wild *Rosmarinus* officinalis was performed to obtain essential oil.



Champor

• **Postbiotics**

Rosemary essential oil

WP3 - Months 6-9 Trial in progress

✓ Water parameters: check water temperature, test pH levels, monitor ammonia levels, measure dissolved oxygen levels.

Abundance

500000

✓ Animal visual inspection: visually inspect animals for signs of disease, injury, or abnormal behavior, observe feeding, record all measurements and observations.

of the fish to adjust the feed quantity (0.8% of the body weight).

	CTRL	REO	AX	РВ
SR (%)	100	100	100	100
SGR (%)	0,7 ± 0,05	0,72 ± 0,03	0,73 ± 0	0,71 ± 0,01

Table 1. Zootechnical indices of trout fed the experimental diets at 45 days of dietary treatment. Analyses are still ongoing.

3-month Feeding trial at Ittica Tranquilli facility, performed on Rainbow trout fed the experimental diets (in triplicate), obtaining four experimental groups



Rainbow trout Oncorhynchus mykiss



All the microencapsulated substances were added to a commercial diet, which was used as control diet, obtaining *four experimental diets*:

1. Ctrl → Control

- 2. REO → ctrl +POP essential oil (2,5 g of POP per kg of feed)
- **3.** AX → ctrl +POP astaxanthin (4.6 g of POP per kg of feed)
- **4.** *PB* → *ctrl* +*POP postbiotics* (12 g of POP per kg of feed)











Future Trial abroad (6 Months)



✓ 6-month feeding trial at Hellenic Center for Marine Research (Greece) will be conducted on gilthead seabream using the same experimental diets (in triplicate)

Gilt-head bream Sparus aurata

✓ Feeding preference test will be carried out using three flavored diets with different synthetic aromas provided by Aquatrade. Each diet will be dispensed through self-

Demand feeder

UNIVPM (DiSVA and D3A)

Future Analyses

- Zootechnical performances
- Real-Time PCR for growth factors, stress response, and welfare;
- Histological analysis on intestine and liver;
- Physical (pH and color) and biochemical analysis (protein, fat content, fatty acid profile and oxidative status) on fish fillets

UNICAM

• Retention capacity of essential oils in the flesh of fish will be







feeders, automatic devices that release feed only when the fish touches a sensor.

\checkmark The data collected will be crucial for determining which aroma the fish prefer,

optimizing the feeding process and reducing both feed and economic waste.

evaluated using SPME coupled with GC-MS.

AMAP "Marche Agricoltura Pesca"

Flavor characterization of fish meat with rosemary: Sensory

evaluation of the flavored trout meat by trained tasters.



