

# Turning wastewater from tuna canning industry into protein-rich biomass through the culture of marine microorganisms

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The fish canning industry generates substantial volumes of wastewater, the proper management of which is crucial for preventing nutrient loss, and minimizing environmental impacts, thus integrating the sector into the circular economy (Cortés et al., 2021).

Brines produced during the cooking stage in canning plants are characterized by their chemical complexity, high levels of organic matter, suspended solids, and salinity (Venugopal et al., 2021). These characteristics hinder the treatment of brines through conventional physicochemical and/or biological methods, resulting in high operational costs and low process efficiency. Bioaugmentation has recently been proposed as an alternative, eco-friendly solution for treating highly saline wastewaters (Anh et al., 2021). This approach involves harnessing brines to cultivate halophilic microorganisms capable of degrading organic matter and utilizing it as nutrients for their growth. However, for this technology to be viable, it is imperative to select microorganisms with key attributes, including active proteolytic activity, rapid growth, and commercial viability.

Thraustochytrids, marine microorganisms naturally involved in decomposing organic matter in aquatic ecosystems, offer significant potential for this technology. Beyond their longstanding industrial use for Omega-3 oil production, thraustochytrids are increasingly recognized as a versatile source of valuable products for the agrifood sector. Moreover, their ability to assimilate waste materials as substrates further enhances their interest (Gupta et al., 2022).

In the above context, this study explores the feasibility of integrating brines from tuna canning plants into the cultivation of an indigenous strain of *Thraustochytrids*, with the aim of producing protein-rich biomass that can be introduced in the value chain of the agrifood sector.

For that, brine samples collected from a tuna canning plant in the Basque Country were chemically characterized after fat removal. Brine was properly diluted with running water and used as culture media after being supplemented only with dextrose (20 g/L) and a F2 medium metal solution (1 ml/L). Cultures were conducted in 5-L fermenters until biomass reached the stationary growth phase. Biomass production was calculated and expressed as volumetric dry weight (g DW/L). Soluble protein (SP), free amino acids (FAA), and phosphate concentrations were analyzed in the residual media and compared with the initial content to determine nutrient removal efficiency (%). Finally, the biomass was chemically characterized and subjected to an enzymatic hydrolysis to produce an L-aminoacid concentrate, as described Romero et al (2018).

Results of the culture process are summarised in the Table 1. Biomass production reached 4.4 g DW L<sup>-1</sup> in a period of 7 days. During the process, 100% of FAA, 28% of SP and 60% of phosphate were effectively removed from the brine-based medium, demonstrating the potential of the strain for bioremediation of saline wastewaters despite the process is not yet optimized. Chemical analysis of algal biomass revealed an organic carbon content of 45.2% DW, 7.9% DW of total nitrogen (TN) and 50.7% DW of protein content. Other primary nutrients present in the biomass were phosphorus (10.29 g/Kg) and potassium (12.17 g/kg) along with several oligo elements. Enzymatic hydrolysis of microbial biomass yielded a liquid hydrolysate composed of 0.59% (w/v) TN and 32 g/L of total amino acids, with 75% (24 g/L) of them being in free form. Originating primarily from the supporting brine, this entirely organic product also included a diverse array of nutritive components released during the hydrolysis process offering a source of nutrients for plant enhancement or other agrifood purposed.

**Table 1.** Main results in terms of process yielding and bioremediation efficiencies, obtained using brine as nutritive medium to cultivate of *Traustochytrium* in 5-L fermenters.

<b>Proces Parameter</b>	
Time to max biomass (Hours)	168
Biomass production (gDW/L)	4.37 ± 0.28
Protein production (g/L)	2.8 ± 0.01
<b>Nutrient Removal Efficiencies</b>	
Soluble protein (%)	28
Free aminoacids (%)	100
Phosphorous (%)	60.6

In summary, this study demonstrates the potential of using brines from tuna canning plants to cultivate Thraustochytrids, yielding protein-rich biomass suitable for agrifood applications. The process effectively removes nutrients from the brine, offering a sustainable solution for managing wastewater in the industry and promoting circular economy principles.

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