OPTIMIZATION OF GROWTH CONDITIONS FOR THE MARINE MICROALGA *TETRASELMIS STRIATA* - AN ALTERNATIVE RAW MATERIAL FOR FISH FEED

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Aquaculture currently faces significant challenges related to the high cost and reduced availability of fishmeal and fish oil for inclusion in the diets of farmed fish. The use of microalgae as an alternative raw material for fish feed presents a great interest as microalgae are an exceptional source of nutrients, such as polyunsaturated fatty acids, proteins and amino acids which can improve fish growth and fillet quality. To improve the microalgal biomass production and increase biomass enrichment in specific nutrients, important functional growth parameters must be optimized.

The marine microalga *Tetraselmis striata* was cultivated in different drilling waters originating from the facilities of Plagton S.A a commercial fish farm located in Western Greece which naturally have different salinities. The waters employed in the present study were taken straight from the drills and presented no nutrient load, thus the supplementation of minerals was essential for microalga's growth. The effects of salinity, pH, and initial nitrogen and phosphorus concentration ratios on the specific growth rate and ability of the strain to biosynthesize proteins, lipids, polysaccharides and pigments were studied. Laboratory-scale experiments were performed under non-aseptic and suspended growth conditions.

T. striata growth was initially examined in drill water with high salinity (38‰) that further was enriched with nitrogen and phosphorous and with or without the addition of trace elements. The results revealed low biomass productivities ranging from 32.2 to 40.0 mg L^{-1} d^{-1} with specific growth rates varying from 0.062 to 0.100 d^{-1} and lipid contents of 9.3-24.0% (Table 1). It was concluded that these low biomass efficiencies were probably due to high salinity and the chemical composition of growth medias suppemented in the 38‰ waters. Consequently, growth of the microalgae was evaluated in less saline drilling waters (29‰) but supplemented with different nutrient media. The waters was enriched with a modified F/2 substrate or the commercial fertilizer Nutri-Leef (composed with 30%-TN, 10% -P, 10% -K) with or without the addition of an inorganic carbon source ($NaHCO_3$). The resulting maximum biomass productivities ranged from 69.3 to 85.0 mg L^{-1} d^{-1} while lipid content was 10.8-13.7% (Table 1). Nutri-Leef 30-10-10 with the addition of inorganic carbon produced the highest biomass yields and was therefore used as the matrix to study the effect of pH. Different pH levels of 7 and 8 were tested and pH 8 presented significant biomass productivity yields (79.81 mg L^{-1} d^{-1} with a specific growth rate of 0.156 d^{-1}) and a maximum oil content of 26.4% (Figure 1).

The biomass of *T. striata* produced under the above optimum growth conditions was rich in carbohydrates, proteins and pigments (36.5%, 38.25% and 3.6%, respectively) (Table 2).

Additionally, the lipids of the microalga biomass contained 10-14% EPA, indicating its high value for incorporation into conventional fish feed.

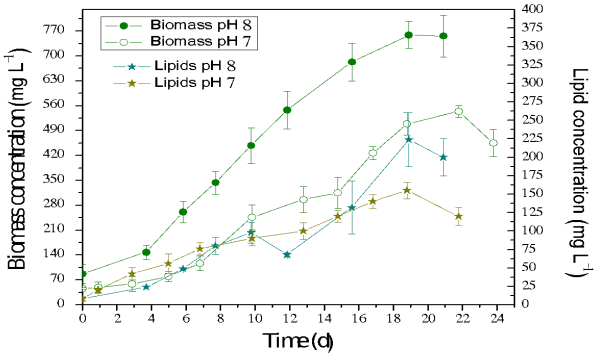

Figure 1. *T.striata* growth and lipid accumulation in Nutri-Leef and $NaHCO_3$ at pH 7 and 8.

Table 1. Results of different growth conditions on *T. striata* biomass.

SUBSTRATE	SALINITY ‰	BIOMASS PRODUCTIVITY mg L^{-1} d^{-1}	SPECIFIC GROWTH RATE d^{-1}	LIPID CONTENT % dw	LIPID PRODUCTIVITY mg L^{-1} d^{-1}	N:P
Mineral medium (N,P)	38	32.2	0.062	24.1	14.0	45
Mineral medium (N,P)	38	40.0	0.100	9.3	6.9	7.4
Mineral medium (N,P) and trace elements	38	38.0	0.096	12.8	12.6	7.4
Modified F/2	29	69.3	0.088	13.7	25.1	46.9
Nutri-Leef	29	70.0	0.132	11.1	9.0	3.0
Nutri-Leef and $NaHCO_3$	29	85.0	0.156	10.8	10.5	3.0

Table 2. Effect of pH level at optimum conditions, on the biomass and growth of *T. striata.*

SUBSTRATE	BIOMASS PRODUCTIVITY mg L^{-1} d^{-1}	SPECIFIC GROWTH RATE d^{-1}	LIPID % dw	CARBOHYDRATE % dw	PROTEIN % dw	PIGMENT % dw
Nutri-Leef $NaHCO_3$, pH=8	79.8	0.156	26.4	36.5	38.3	3.6
Nutri-Leef $NaHCO_3$, pH=7	60.12	0.135	25.6	16.1	35	3.8

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