SEMI-CONTINUOUS CULTURE OF *Nannochloropsis oculata* IN OPEN SYSTEMS AND NITRATE CONCENTRATION EFFECT

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Introduction. In recent years, biotechnological industry of photosynthetic microorganisms has grown and become diversified. However, their commercial production has been limited by high production costs (1). In order to reduce the production costs, several efforts are being made by developing efficient photobioreactors (2). Therefore, the use of open culture systems, using solar energy is an economical alternative of algal biomass production. Moreover, a large-scale outdoor production of microalgae should consider these critical issues: biotic factors such as pathogens (bacteria, fungi, viruses) and competition with other microalgae; abiotic factors such as light (quality, quantity), temperature, O_2, CO_2, pH, salinity, and nutrient concentration (3) and operational factors such as shear stress by mixing, depth, and harvest frequency or dilution rate. Concerning to harvest frequency, there are few studies about this subject in outdoor systems.

Thus, the objective of this work to establish the effect of semi-continuous culture and feeding nitrate between harvests in the production of algal biomass in open systems.

Methods. *Nannochloropsis oculata* strain was grown in f/2 media (4) in 200 L open systems under greenhouse conditions during 70 days and 5 harvests. Light irradiance, temperature of the environment and within the culture were recorded every 10 min with a T&D Corp RTR-500 data logger. Systems were harvested by removing half of the volume and replacing it with both fresh media (RW1), and fresh medium supplemented with the nitrate lost in the spent media (final concentration 250 mg/L) (RW2). Harvesting cycles took place when nitrate concentration reached roughly 50 mg/L. Biomass (dry weight), nitrates, pH and lipids (cold extraction with hexane and gravimetric determination) were carried out during the kinetics.

Results. *N. oculata* growth was followed by the consumption of nitrates, apparently without the limitation of nitrate except for the second cycle, in which nitrogen limitation was achieved at 27th day (overall culture) (Fig. 1). The pH interval was about 8 at the beginning of each cycle, and about 10 at the end. For the last three cycles, higher biomass production was observed in the system with higher nitrate level, reaching 0.98, 1.16, 1.26 g/L, respectively (Fig. 1). These values represent an increase of 14.6 % ± 1.5 compared with the system of lower nitrate level. In both systems, as systems were being harvested, the new cycles showed an increase in final biomass. However, the effect was not evident in the growth rate raise (Table 1).

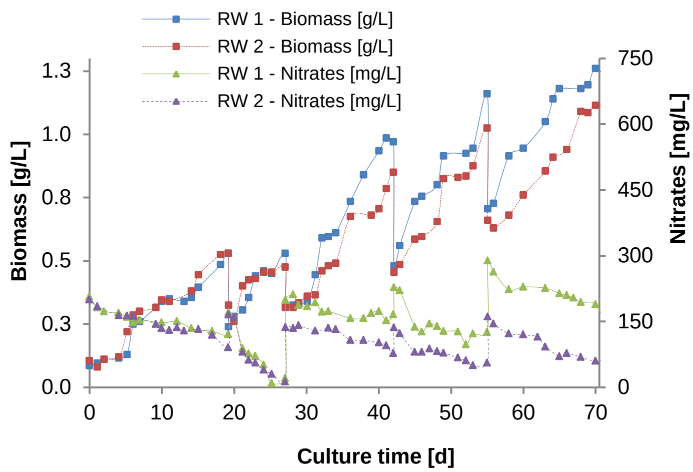

Figure.1 Semicomtinuos culture and nitrate concentration effect on biomass production of *N. oculata,* growth in open systems.

Table 1. Parameters for each harvesting cycle during the culture of *N. oculata* in open systems.

Harvesting cycle	Culture time [d]	μ_{max} [d^{-1}]		Temp. interval [°C]	Max. irradiance [μmol photon /m^2 s]
		RW 1	RW 2		
1	19	5.51	4.81	6.6 – 36.3	780
2	8	4.84	4.23	6.2 – 48.0	843
3	15	5.55	5.08	9.7 – 38.6	917
4	13	5.58	5.32	10.3 – 39.5	807
5	15	4.32	3.85	8.1 – 39.2	873

Conclusions. The systems in which nitrate was restored to 250 mg/l, showed higher biomass production (growth). Semi-continuous culture allows for each harvest (or recultivation) an increment in the biomass.

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