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Outdoor mass microalgae production in Bahia Kino, Sonora, NW Mexico

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Abstract

The diatom *Chaetoceros muelleri* was grown in outdoor mass cultures under the winter, spring and summer conditions of Bahia Kino, Sonora, NW Mexico. The solar irradiance in winter was close to 60% of that available in spring and summer, but the cell concentrations and the harvestable biomass were one order of magnitude higher in spring and summer than in winter. There was no difference between the biomass harvested after 2 and 3 days in winter and summer, whereas in spring it was higher after 3 days. The protein content was significantly lower in winter, and carbohydrates and lipids were higher in winter and spring. The number of cells and the amount of harvestable biomass of outdoor cultures of *C. muelleri* depend on the temperature prevailing in each season, which causes significant differences in growth rates and in biochemical composition.

Keywords: *Chaetoceros muelleri*; Outdoor cultures; Light utilization; Growth efficiency; Biomass production; Proximate composition

1. Introduction

Microalgae are considered the best food source in commercial hatcheries, which have daily requirements of several m^3 of cultures. In tropical and subtropical countries almost all commercial-scale algal cultures are grown outdoors that, in view of seasonal environmental changes, may cause variations in growth, in production and in composition (López-Elías et al., 2005).

In this work, we determined the efficiency of light utilization of outdoor cultures of the diatom *Chaetoceros muelleri* grown under winter, spring and summer conditions, in a commercial hatchery located in Bahia Kino, Sonora, NW Mexico (28°50′N; 111°56′W).

2. Material and methods

The strain of the diatom used (CCMP 1316 = CHGRA) is the most popular in Mexican commercial hatcheries. It was grown indoors at 25 °C and with 9.5 E m^{-2} day^{-1} photon flux, using the traditional multi-step procedure (cultures of progressively increasing volumes, of 0.1, 2, 5, 16 L; air bubbling was provided starting at 5 L). The growth medium was f/2

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Table 1
Number of light and dark hours (L:D). Age of cultures (days). Total irradiance ($\sum$ (E m^{-2})) and accumulated temperature ($\sum$ (°C day)) in 2 or 3 days. Mean cell concentrations (*N*: in 10^6 cells mL^{-1}) and cell production for accumulated degree-day and accumulated irradiance (10^3 cells $°C^{-1}$ and 10^3 cells E^{-1}) of outdoor cultures of *Chaetoceros muelleri* in different seasons

Season	L:D (hours)	Age	$\sum$ (E m^{-2})	$\sum$ (°C day)	*N*	10^3 cells $°C^{-1}$	10^3 cell E^{-1}
Winter	10.5:13.5	2	118	38.8	0.49 (0.03)	12.73	4.19
		3	182	58.2	0.95 (0.05)	16.24	5.19
Spring	11.5:12.5	2	196	49.3	3.70 (0.02)	75.05	18.88
		3	287	74.0	4.15 (0.04)	56.08	14.46
Summer	12.5:11.5	2	200	61.1	3.44 (0.01)	56.25	17.19
		3	315	91.7	3.59 (0.01)	39.15	11.40

(Guillard, 1975), prepared with industrial-grade reagents.

A mixture of six 3 day-old cultures was used to start six 300 L transparent (>90% transmittance) fiberglass cylinders (1.5 m high, diameter 0.5 m), with initial cell concentrations of 0.2×10^6 cells mL^{-1}, that were maintained under natural conditions in winter, spring and summer. These experiments were performed three times in each season. Irradiance above the cultures and water temperature were measured hourly and pH was maintained nearly constant at 8 ± 0.5 with unmetered CO_2 injection during light hours.

After 42 and 72 h, samples of each culture were used to determine the respective cell concentrations and three 50 mL aliquots were concentrated on Whatman GF-C glass fiber filters of known dry weight, washed with 5 mL of ammonium formate to eliminate sea salts and dried to constant weight at 60 °C to obtain the total dry mass (DW). The inorganic content (AW) was determined after ashing at 475 °C for 12 h, and the organic mass (OW) was calculated as the difference between DW and AW.

Other samples, obtained in triplicate for each culture and type of analysis, were concentrated on Whatman GF-C filters and used for protein, carbohydrate and lipid determination following Lowry et al. (1951), Dubois et al. (1956) and Pande et al. (1963) following the procedures detailed in López-Elías and Voltolina (1993).

The mean accumulated irradiance ($\sum$ (E m^{-2}): mean of the total irradiance received during 2 and 3 days by each culture, calculated by integration of the hourly readings) and the accumulated temperature ($\sum$ (°C): sum of daily mean water temperatures) were used to calculate the efficiency of cell and biomass production for each season.

All data were normal and homoscedastic. The mean values of the total harvestable biomass, the individual dry weights and the protein, carbohydrate and lipid contents of each date and season were compared by two-way analysis of variance (Zar, 1996).

3. Results

The number of cells produced for each accumulated °C ranged from 12.7 to 75.1×10^3 cells mL^{-1}. It was between 2.4 and 5.9 times higher in spring and summer, and the best cell production was in spring. A further seasonal difference was that in winter the efficiency of production increased after day 2, whereas it decreased in spring and especially in summer (Table 1).

The total irradiance received by the cultures in winter was close to 60% of that available in spring and summer, but the cell concentrations observed in these two seasons were between four and seven times higher than those of the winter cultures. This shows that, because of the low temperature, the efficiency of utilization of light energy in winter ranged from 4.2 to 5.2×10^3 cells mL^{-1} E^{-1} after 2 and 3 days. In comparison, the light utilization efficiencies varied between 11.4 and 18.9×10^3 cells mL^{-1} E^{-1} during the warmer seasons (Table 1).

The harvestable biomass and the culture growth rates were lower in winter and there were no appreciable differences between spring and summer after day 2. After 3 days the accumulated irradiance was 10%

Table 2
Culture age (days). Total harvestable dry weight (TDW: mg L^{-1}). Mean growth rate (divisions day^{-1}) and individual organic weight (OW) of outdoor cultures of *Chaetoceros muelleri* in different seasons. Standard deviations in parenthesis. Equal letters indicate lack of significant differences (two-way ANOVA, $\alpha = 0.05$: $a < b < c$)

Season	Age	TDW	Div. day^{-1}	OW
Winter	2	31.5 (1.6) a	0.65 (0.037)	6.37 (0.59) b
	3	38.0 (2.2) a	0.96 (0.046)	4.07 (0.35) b
Spring	2	104.0 (27.0) b	2.10 (0.011)	2.80 (0.57) a
	3	195.0 (33.0) c	0.17 (0.003)	3.34 (0.70) a
Summer	2	114.0 (40.0) b	2.05 (0.006)	3.31 (1.15) a
	3	145.0 (32.0) b	0.06 (0.002)	3.74 (0.78) a