Mixotrophic cultivation of the marine microalga *Nannochloropsis salina*: effects of glycerol and CO_2 on growth and biochemical composition

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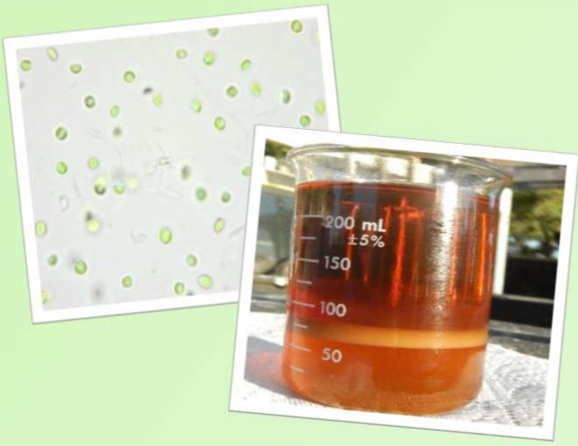
INTRODUCTION:

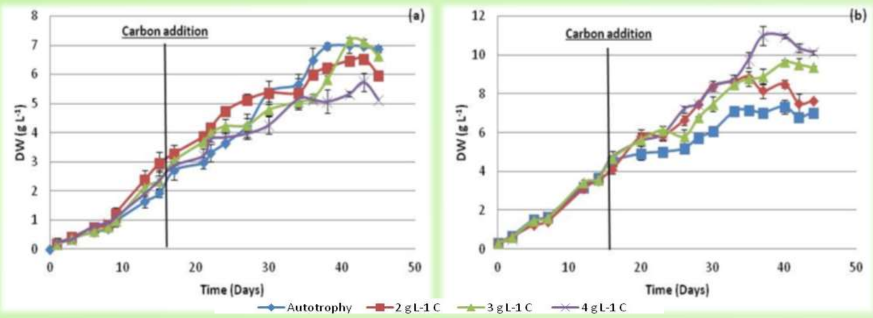
Nowadays it is increasingly recognized the importance that microalgae biomass can play in several sectors. Currently photoautotrophic cultivation is the main modality of cultivation but it's limited by light availability and CO_2 supply, with high production costs. Nutrients and CO_2 are two of the major costs and then it becomes necessary to find cheapest modalities of cultivation in order to make more economically sustainable the whole system.
Mixotrophic cultivation using raw materials rich in nutrients and carbon could become an alternative way to obtain high biomass production, recovering C-rich waste.

AIM:

The aim of this work has been focused on the possibility to use glycerol, a C-rich by-product coming from biodiesel production, as carbon source for the mixotrophic growth of the marine microalga *Nannochloropsis salina.* Microalgae was mixotrophically grown in 0.5 L photobioreactors (PBRs) with glycerol at different concentrations (2-3-4 g L^{-1} of C) with and without CO_2 supply.



Nannochloropsis salina growth: (a) growth curve without CO_2 supply; (b) growth curve with CO_2 supply



Biomass composition of *Nannochloropsis salina* grew on glycerol with and without CO_2 supply

Strains	CO_2 supply	Carbon (g L^{-1})	Lipid content (% DW)	Protein content (% DW)	Carbohydrates content (% DW)
Nannochloropsis salina	+	0	35.6 ± 0.2a	13.3 ± 0.7a	17.7 ± 0.8b
	-	2	34.6 ± 0.3a	13.1 ± 0.1a	16.1 ± 0.1b
	-	3	45.7 ± 1.8b	14.1 ± 0.2a	5.2 ± 0.7a
	-	4	44.8 ± 3.4b	12.3 ± 0.6a	6.3 ± 1a
	+	0	35.5 ± 2.1a	12.5 ± 0.9a	18.9 ± 2b
	+	2	34.4 ± 1.7a	13.7 ± 0.5a	18.5 ± 1.4b
	+	3	46.6 ± 1.8b	16.3 ± 0.6b	6.5 ± 0.5a
	+	4	42.5 ± 1.1b	13.8 ± 0.5a	7.5 ± 0.7a

Values in the same column followed by the same letter are not statistically different at $p < 0.05$ according to Tukey test.

RESULTS:

Nannochloropsis salina grew mixotrophically with success at all concentrations provided.
In particular when 3 g L^{-1} of C were provided, the biomass dry weight DW was 7.33±0.35 and 10.98±0.50 g L^{-1} for autotrophy and mixotrophy, respectively. The biomass composition analysis showed that lipid content in mixotrophy was higher than in autotrophy, particularly when 3 g L^{-1} of C were provided (35.5±2.1 and 46.6±4.8 % of DW for autotrophy and mixotrophy, respectively).
In the mixotrophic trials where CO_2 wasn't supplied, we observed that the recorded growths were very similar between the mixotrophic and autotrophic PBRs (similar DW of about 7 g L^{-1}). However, also in this case, total lipid content in mixotrophy was upper than in autotrophy, (35.6±0.2 and 45.7±1.8 % of DW for autotrophy and mixotrophy, respectively).

CONCLUSIONS:

Result of this work indicated that *Nannochloropsis salina* can mixotrophically grow on glycerol both with and without CO_2 supply. Glycerol has been suitable replacing completely CO_2-C when this latter was omitted, showing algae biomass production similar to those obtained in autotrophy, producing had a final biochemical composition better to those of algae grew in photoautotrophic condition.

This study is part of DANCE project , financed by **Fondazione Cariplo (2014-0587)** has the aim to recover renewable carbon and nutrient sources from lombardy agro-industry to be used to produce microalgal biomass rich in high added-value molecules by developing mixotrophic bio-technology.