

Heterotrophic Culture of *Chlorella protothecoides* in Various Nitrogen Sources for Lipid Production

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Abstract The influences of urea, yeast extract, and nitrate as the nitrogen source on heterotrophic growth of four strains of *Chlorella protothecoides* were investigated in 9-day feed-batch cultures. Biomass dry weight concentration (DWC) and lipid yield (LY) of the four strains in all media were compared. The highest LY in 9 days was 654 mg/L/day by UTEX 255 in 2.4 g/L KNO₃ medium with a biomass DWC of 11.7 g/L and lipid content of 50.5%. Using green autotrophic seeds instead of yellow heterotrophic seeds improved the biomass DWC (13.1 vs. 11.7 g/L), LY (850 vs. 654 mg/L/day), and lipid to glucose consumption ratio (0.607 vs. 0.162). Moreover, 17.0 g/L DWC and 489 mg/L/day LY were obtained from the sequentially mixed-nitrogen medium, and the lipid to glucose consumption ratio was improved to 0.197 from 0.162 in 2.4 g/L nitrate medium and from 0.108 in 4.2 g/L yeast extract medium in the first batch.

Keywords Algae · Biofuel · Nitrogen · *Chlorella protothecoides* · Lipid

Introduction

As energy prices reach historical highs and environmental concerns continue to grow, broad interest exists in producing and utilizing biofuels from domestic biomass resources. Algae, a group of organisms that can grow, autotrophically or heterotrophically, in freshwater or saltwater are one such resource. The potential for producing biofuels (especially biodiesel) from algae was demonstrated and extensively studied about 20 years ago [1]; on the basis

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