



Lipid production by *Rhodospiridium toruloides* Y2 in bioethanol wastewater and evaluation of biomass energetic yield

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HIGHLIGHTS

- ▶ *R. toruloides* Y2 was cultured in bioethanol wastewater for lipid production.
- ▶ Biomass energetic yield was evaluated by biomass, lipid content and substrate COD.
- ▶ A method was chosen to accelerate lipid production and avoid catabolite repression.

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ABSTRACT

The oleaginous yeast *Rhodospiridium toruloides* Y2 was employed to remove waste nutrients from bioethanol wastewater while simultaneously producing biomass enriched in microbial lipids. Under optimal conditions, the COD degradation ratio, biomass and lipid content reached 72.3%, 3.8 g/l and 34.9%, respectively. For accelerating biomass and lipid accumulation, different feeding strategies of substrate were conducted. The biomass and lipid production increased by 39.5% and 53.8%, respectively, when glucose at 1.2 g/(l d) was added during the last three days of the cultivation. An equation was established to estimate biomass energetic yield. Under optimal conditions, the biomass energetic yield was 50.9% and an increase of 26.0% was obtained by feeding glucose at 1.2 g/(l d) during the last three days. The fatty acid composition of the lipids was similar to that from plant oils and other microbial lipids, and could thus be used as raw material for feed additives and biodiesel production.

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1. Introduction

The bottle neck for the industrialization of biodiesel is the high cost of its raw material which accounts for 70–85% of total cost (Yousuf et al., 2010). Microbial lipids are being explored as raw materials of the production of biodiesel and functional oils (Minkevich et al., 2010; Papanikolaou and Aggelis, 2011). An important advantage offered by the application of the oleaginous microorganisms is their ability to produce lipids from waste organic matters. Consequently, to optimize the cost of the process, as well as to increase its environmental benefit, some waste materials had been studied as feedstock for single-cell oil production by fermentation with oleaginous microorganisms (Chi et al., 2011; Chinnasamy et al., 2010; Pittman et al., 2011; Xue et al., 2010), but more waste streams remain to be investigated for cost-effective microbial lipid production.

Wastewater from bioethanol manufacturing is difficult to treat (Li et al., 2009). Although upflow anaerobic sludge blanket (UASB)

and aerobic activated sludge process can remove 99% of chemical oxygen demand (COD), five-day biochemical oxygen demand (BOD₅) and suspended solids (SS) and produce biogas (Liu et al., 2011b), they require start-up periods of up to 3 months and produce sludge (Liu et al., 2011a). Therefore, it is desirable to develop an efficient and economical treatment approach for bioethanol wastewater.

Therefore, the present study aimed to: (1) recycle the energy contained in bioethanol wastewater and transfer it into lipid-enriched cells by an oleaginous yeast *Rhodospiridium toruloides* Y2, (2) develop a method to evaluate the energy yield of the fermentation, (3) investigate the effects of different glucose feeding strategies on the treatment of the wastewater, (4) analyze the fatty acid composition of the lipids to estimate their potential application for the production of biodiesel and feed additives.

2. Methods

2.1. Organism, media and chemicals

Yeast extract (containing 3.0% ammonium-N and 9.0% total nitrogen) and peptone (containing 3.0% ammonium-N and 14.5%

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