PhD Open Days

Direct lipid and carotenoid extraction from *Rhodosporidium toruloides* broth culture after high pressure homogenization cell disruption: strategies, methodologies, and yields.

PhD program in Biotechnology and Biosciences

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Introduction

Biodiesel production from oleaginous microorganisms requires several steps: biomass production, cell disruption and lipid extraction, followed by a transesterification reaction. The lipid extraction step is a crucial step since it will affect the biodiesel quality, quantity and the cost efficiency of the process. Post harvesting process techniques such as cell disruption methods (ultrasound, milling, high pressure homogenization (HPH), etc.), usually used to improve lipids extraction from the cells, are required to obtain an economically feasible biodiesel production process.

Among the different cell disruption techniques, HPH is easily scalable and considered one of the most environmentally safe methods: HPH can be performed immediately after biomass harvesting and it is not necessary the addition of any chemical products. Afterwards, the lipids and carotenoids can be extracted using different solvents that can access easily to the cell content.

Aims

The aim of the present work is to develop a protocol combining a cell disruption technique and solvent

Results

Effect of High Pressure Homogenization on *R. toruloides* cell disruption



Figure 1 – a-d – FSC *versus* SSC dot plots concerning *Rhosdosporidium toruloides* cells before and after the three cycles of High Pressure Homogenization (HPH) at 600 bar. Two populations exist, A corresponding to intact *R. toruloides* cells which kept their structure, size and internal complexity (no FSC/SSC signal alteration) and B composed of medium particles and HPH damaged *R. toruloides* cells which are smaller and empty with less FSC/SSC signal; e – Cell rupture percentage obtained after each of the three HPH passages.

extraction to co-extract lipids and carotenoids from *Rhodosporidium toruloides* biomass, directly from the broth

culture, without using any previous either harvesting or dewatering steps. The results obtained were compared

with the results attained using a conventional lipid extraction technique.

The process here presented is an easy, simple, inexpensive and environmentally friendly methodology

involving the simultaneous extraction of carotenoids and lipids directly from *R. toruloides* broth culture which

has never been reported before and can greatly improve the economics of the biodiesel production process.

Methodology



Effect of HPH on lipids and carotenoids extraction

For the HPH treated yeast biomass it was possible to obtain a higher lipid recuperation yield, when compared to the yeast biomass without HPH treatment.

Only a small part of the lipids and carotenoids of *R. toruloides* were released to the extracellular medium after HPH.

In all the samples, the most representative fatty acids were palmitic acid (~25 % w/w DCW of TFA), oleic acid (55 % w/w DCW of TFA) and linoleic acid (~10% w/w DCW of TFA).

Direct hexane extraction of the broth culture after HPH is an easy, simple and inexpensive method to obtain lipids and carotenoids rich extracts saving time, chemicals and energy comparatively with conventional methods.

Sunflower oil was also used to perform a liquid-liquid extraction directly on the broth culture and on the supernatant obtained after the HPH treatment since it is a low-cost safe product, usually used in the food and cosmetic industries, and its usage can avoid the use of less human safety solvents such as hexane.

Conclusions

This work investigated, for the first time, the use of high pressure homogenization, followed by an extraction solvent protocol, to simultaneously extract lipids and carotenoids from *Rhodosporidium toruloides* biomass, directly from the broth culture, without using any harvesting technique. The results obtained were better than those obtained using conventional lipid extraction techniques. The process here presented is an easy, simple and inexpensive method to obtain lipids and carotenoids rich extracts with high economical value, using low cost and environmentally friendly chemicals. This process uses adequate solvents for the food industry and can be easily performed with other microbial biomass (other yeasts, bacteria, and microalgae).

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