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Rhodotorula graminis Lipids Biosynthesis from renewable carbon sources for the production of biodiesel

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Abstract

In recent years, industrial fermentation studies have been focused on processes for biofuels production that can move the market dependence away from fossil based energy sources. In order to avoid the competition with food crops, process studies have moved from the first generation to the so called second generation biofuels, that would be manufactured from agricultural and forestry residues and from lignocellulosic non-food energy crops.

Sugars obtained from lignocellulosic feedstock are a good carbon source for oleaginous microorganisms, that can produce lipids definitely comparable to vegetable oils, thus suggesting their use in fermentation processes for the production of second generation biodiesel. Moreover, in accordance with the recent increase of vegetable oils cost, the use of microbial oils offers additional economical advantages.

Among oleaginous yeasts, *Rhodotorula graminis* is known for its ability to accumulate large amounts of intracellular lipids, which may achieve up to the 70% of the total cellular dry weight.

This study describes how the yeast *Rhodotorula graminis* DPVG 4620 could be a good candidate for converting renewable feedstock into lipids, due to its ability of growing on different carbon sources, such as lignocellulose-derived C6 and C5 sugars, and to its resistance to the most representative inhibitors formed as by-products in the hydrolysis of lignocellulosic biomass (i.e. furfural, HMF, acetic acid).

The *Rhodotorula* performance has been investigated in a scale ranging from 100 ml shake-flasks to 20 liters fermentor, and the more suitable culturing conditions to achieve high yields of lipids and biomass have been identified.

In particular, fermentation experiments with glucose as sole carbon source, or with pulse additions of glycerol, were assessed on a 1-liter scale reactor. In the first case, a final lipid content of 45% g $g_{d.w.}^{-1}$ and a yield of 0,17 g $g_{Csource}^{-1}$ were obtained after 67 hours of fermentation, while a lipid content of 61% g $g_{d.w.}^{-1}$ with a yield of 0,26 g $g_{Csource}^{-1}$ were obtained by adding glycerol.

The fermentation scale up was performed in a 20 liters stirred tank reactor, in which a fed-batch process was settled with a glucose based medium, by this way very high cellular densities and lipid concentrations were obtained. A mixture of xylose and glucose (1:1 ratio) was also tested in fed-batch mode, showing an overall good performance of the yeast but with a lower lipid productivity.

Further experiments were carried out in a 1-liter fermentor with a mixture of sugars to which furfural, HMF and acetic acid were added, in order to simulate a real dilute-acid spruce hydrolysate. Even in this case *R. graminis* has reached very high yields, showing its good potential despite the presence of inhibitors.