

Study on fermentation capacity of several microbial strains for bioethanol production from lactose

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The overall objective of the work was to select microorganisms able to ferment lactose and produce ethanol in batch system from 12 microbial strains available in our culture collection

Experimental:

Kluyveromices marxianus DSM 5418, *Kluyveromices lactis var.lactis* DSM70799, *Zymomonas mobilis* DSM 424, *Saccharomyces cerevisiae* 203, *Saccharomyces cerevisiae* 17/17, *Saccharomyces cerevisiae* SMR-4 Killer, *Saccharomyces cerevisiae* K5-51 Rhe-, *Saccharomyces cerevisiae* K5-5a Spencer, *Saccharomyces cerevisiae* Hansen CBS 5926- Boulardii, *Saccharomyces cerevisiae* Crystal Japan, *Saccharomyces cerevisiae* Cara R581, and *Saccharomyces cerevisiae* CMIT21 microbial strain from the collection of microorganisms CMIT from our University were the microbial strains employed in the experiments.

After incubation at 30°C in a shaker at 150 r.p.m. for 24 hours, the obtained cultures are used as inoculums for fermentation medium containing lactose as carbon and energy source. As fermentation medium, we have used a modified version of Y.P.D. medium by replacing dextrose 2% with lactose 10% as carbon and energy source. Accordingly, the fermentation medium formula used in this study is: 1% yeast

extract, 2% casein peptone and 10% lactose, and pH was adjusted in the initial condition to reach 5.6-5.8.

Fermentation

Fermentation was performed in triplicates for each strain using 15 ml plastic test tubes covered with screw caps in which injection needles are inserted. The tubes containing inoculated lactose media were incubated at 30°C in a water bath and samples were collected every 12 hours, centrifuged at 1000 rpm for 30 minutes and the supernatant was frozen until analysis.

Analytical Methods, material and equipment

Ethanol concentration was estimated by NIR methods using AlcoLyzer M [Anton Paar – Austria]. This device uses a patented method (US 6,690,015; AT 406711) based on near infrared (NIR) spectroscopy to determine the alcohol content in a highly alcohol-specific range between 1150 nm and 1200 nm. The evaluation method uses the significant alcohol peak in this area and two spectral points very close to it for defining the baseline.

Results and Discussions

Total lactose concentration decreased and ethanol concentration increased with increasing time and fermentation was completed in 72 hours in all experiments. The experimental data, respectively alcohol concentration in %v/v, analyzed by NIR method using AlcoLyzer M (Anton Paar) were calculated. Fermentation efficiency (FE) varies between 3.5 - 71%, the best results are obtained for *Kluyveromices marxianus* DSM 5418 (70.86%) and *Kluyveromices lactis var. lactis* DSM70799 (65.75%).

Conclusions

For alcoholic lactose fermentation the best results were obtained with *Kluyveromyces marxianus* DSM 5418 and *Kluyveromyces lactis var.lactis* DSM70799 that produced alcohol solutions with concentrations between 4.50 - 4.85% (v / v) and achieved yields of 71-66% reported to lactose content of fermentation medium. These two yeasts will be used in future studies for alcoholic fermentation of lactose from whey obtained after cheese separation.

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