

TITLE: LACTIC ACID BACTERIA: OVERVIEW OF CONTAMINATION IN BIOETHANOL INDUSTRY

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ABSTRACT:

Brazil is one of the largest bioethanol producer of the planet, producing over 30 billion liter per year, corresponding to around 1/3 of global production. Brazilian distilleries use sugar molasses and sugarcane juice as substrates for fermentation process that uses *Saccharomyces cerevisiae* strains. Bacterial contamination is an ongoing problem for commercial bioethanol production, since bacteria may compete with yeasts for nutrients becoming a constant drain on carbon available to conversion into ethanol. Furthermore, the contaminants can produce byproducts that are inhibitory to yeast during the ethanol production, reducing the productivity and the profitability of the process. Lactic acid bacteria (LAB) are very significant contaminant in bioethanol production process and their expansion has indeed a negative effect in the process causing serious economic losses. Hence, it is critical to have a better understanding of incidence and diversity of them in bioethanol production process. This study aimed to investigate the diversity of lactic acid bacteria (LAB) contamination present in different Brazilian distilleries of bioethanol production. 33 LAB isolates were obtained from 20 distinct distilleries in 2015/2016 harvest season using MRS Agar medium. To identify the isolates, genomic DNA were extracted and amplified by PCR 16S rRNA using p10f and p1100r primers. The amplification products were purified and sequenced using ABI3500XL Series (Applied Biosystems). We found 8 different OTU (Operational Taxonomy Unit). Most of isolates were identified in the genus *Lactobacillus* (90,1%). *L. paracasei* was the most widespread LAB as it was found in 70,0% of industrial units assessed, followed by *L. fermentum* and *L. ferruginis*, both present in 15,0% of them. We also discovered *L. amylovorus* (5,0%) and *L. harbinensis* (5,0%). Likewise, *L. fermentum* and *L. paracasei* were found at the same distillery (5,0%). Besides, we found *Staphylococcus* sp. (5,0%) and *Acetobacter* sp. at the same industrial unit. The results show clearly that LAB diversity is an important component of bacterial contamination in the bioethanol fermentation process due to their ability to grow in harsh environmental conditions. Additionally, these results support the overall accepted belief that LAB, particularly *Lactobacillus* sp., are the most prevalent contaminants of bioethanol production process, although some microbiome differences may possibly due to endemic infections.

Keywords: bioethanol, contamination, diversity, fermentation process, lactic acid bacteria

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