Nisin Activity Against Contaminant Bacteria Isolated From Bioethanol Fermentation Tanks

Natalia Maia (I), Rachel Rigotti (I), Renata Silva (I), Bruna Lange (I), Giuseppe Meca (II), Renata Macedo (I), Fernando Luciano (I)

(I) PUC - PR - Pontifícia Universidade Católica do Paraná (BR 376 Km 14, São José dos Pinhais, PR, Brazil 83010-500), (II) UV - ES - University of Valencia (Av. Vicent Andrés Estellés s/n, 46100 Burjassot, Spain)

Resumo

Nisin is a bacteriocin (lantibiotic) produced by Lactococcus lactis, which is widely used in the food industry as a natural antimicrobial to avoid the growth of pathogenic and spoilage bacteria. The objective of the present work was to evaluate the antimicrobial activity of nisin towards Leuconostoc mesenteroides and Lactobacillus fermentum, which are common contaminants in Brazilian bioethanol production plants. These bacteria are normally present in the wort and significantly reduce the sugar-to-ethanol conversion capacity of Saccharomyces cerevisiae. Firstly, the Minimum Inhibitory Concentration (MIC) of nisin was determined for each one of the bacteria in tubes containing CSN broth (supplemented sugarcane broth). Then, a wide range of nisin doses was tested (0.125 – 7,500 ppm) during CSN (10 oBRIX and pH 5.0) fermentation. The broth was co-inoculated with Saccharomyces cerevisiae, Lactobacillus fermentum and Leuconostoc mesenteroides and incubated for 24 h at 30oC. Then, samples from each batch were submitted to microbial count, oBRIX analysis, yeast viability and determination of ethanol content. All tests were performed 3 times in duplicate (n=6). The MIC for both bacteria was 0.75 ppm. The average bacterial population in the control group (non-treated samples) was 10.4 log UFC/mL after the fermentation period. The lowest dose used, 0.125 ppm, did not present significant reduction of the bacterial contamination. However, all other doses presented a drastic of the bacterial population: 4.3 log UFC/mL with 0.375 ppm; 5.8 log UFC/mL with 0.75 ppm.
ppm (=CIM), 5.7 log UFC/mL with 7.5 ppm, 7.5 log UFC/mL with 750 ppm; and > 9.4 log CFU/mL with 7,500 ppm. None of the doses tested presented a significant reduction of Saccharomyces cerevisiae population, and the yeast viability was always > 95%. Groups treated with $\geq 0.75$ ppm presented lower o BRIX values and higher ethanol concentration than the control group. These results show that nisin may be used as a natural antibacterial agent during the production of bioethanol in order to reduce the population of Lactobacillus fermentum and Leuconostoc mesenteroides, and, consequently, increase the sugar-to-ethanol conversion rate by Saccharomyces cerevisiae.

**Palavras-Chave:** Bioethanol, Natural Antimicrobial, Bacterial Contamination, Bacteriocins

**Agência de Fomento:**