TITLE: ANTIMICROBIAL ACTIVITY OF ORANGE ESSENCIAL OIL ON MICROORGANISMS OF BIOETHANOL PRODUCTION.

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ABSTRACT:

Contamination control in the fuel ethanol industry is carried out with acids and antibiotics, which are often inefficient, generating high production costs and directly affecting the environment. Residues of citrus, Orange Essential Oil (OEO), contain bioactive compounds with antimicrobial activity, having a potential biotechnological interest. The objective was to evaluate the antimicrobial effect of OEO on microorganisms of ethanol fermentation, such as the contaminating bacterial *Lactobacillus fermentum* and *Lactobacillus plantarum* and the yeasts *Dekkera bruxellensis* (contaminant) and *Saccharomyces cerevisiae*. OEO concentrations ranging from 0 to 16000 mg/L, previously prepared, were filtered on 0.45µm porosity membrane, diluted in distilled water and homogenized with Tween 80 (1:1). Yeast inoculants (*D. bruxellensis* and *S. cerevisiae*) were prepared in 125 mL erlenmeyers containing 50 mL of 4°Brix cane broth and placed in shaker at 30ºC/160rpm/24 hours. After which the counting was done in Neubawer's chamber, standardizing the inoculum ate 10^7 cells/mL. For the bacterial inoculum the same procedure was done, but the standardization was reading in spectrophotometer (absorbance at 540 µm) to the value of 0.4. The inoculated falcon tubes (in triplicate) were shaken at 30ºC/160rpm of stirring and a 0.1 mL sample were analyzed at 0, 24, 48 and 72 hours. The growth of the yeast was evaluated by counting in Neubawer's chamber and for the bacteria by the drop-plate method. The OEO characterization was done by Chromatography Gas Mass Spectrophotometry (CG-MS). For *S. cerevisiae*, there was na increase in the lag phase of growth from the concentration of 2000 mg/L, without causing cell death at any of the concentration tested. For *D. bruxellensis* cell death was observed at the same concentration. For bacteria, OEO showed a less pronounced effect on growth, more significant from the concentration of 8000 mg/L. For *L. fermentum* the effect was lower, causing only an increase in the lag phase of growth (8000 mg/L). For *L. plantarum* OEO caused cell death with a reduction of 2 log cycles (8000 mg/L). The use of OEO proved to be efficient in controlling the growth of microorganisms contaminating the ethanol fermentation, and the lowest concentration tested had an antimicrobial effect. In these concentrations the effect on *S. cerevisiae* was much lower, demonstrating selectivity, which is an important characteristic in the context of ethanolic fermentation.

Keywords: orange waste, bioactive compounds, antimicrobial, ethanol fermentation.

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