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### **Minimum in vitro bactericidal concentration of essential oils of oregano (*Origanum vulgare*), pink pepper (*Schinus terebinthifolia*), and melaleuca (*Melaleuca alternifolia*)**

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Concern about health and the search for a better quality of life has grown substantially over the years. From this perspective, the use of natural preservatives, rich in bioactives, has gained prominence among consumers. Thus, the objective was to evaluate the minimum bactericidal concentration (CBM) of the essential oils Melaleuca (*Melaleuca alternifolia*), Oregano (*Origanum vulgare*), and Pink pepper (*Schinus terebinthifolia*). The research was carried out at the Microbiology and Immunology Laboratory of the Biology Department of the Federal Rural University of Pernambuco - UFRPE. Bacterial isolates were used at a concentration of  $3.5 \times 10^7$  CFU/mL (0.5 on the McFarland scale) of *Staphylococcus aureus* (ATCC 25923), and *Escherichia coli* (ATCC 25922) obtained commercially from the American Type Culture Collection. The essential oils were serially diluted in 14 different concentrations (10,000, 5,000, 2,500, 1,250, and 1,000  $\mu\text{g/mL}$  to 500, 250, 125, 62.5; 31.25; 15.62; 7.81; 3.90 and 1.95  $\mu\text{g/mL}$ ) in dimethyl sulfoxide. Then, 100  $\mu\text{l}$  of these solutions were added to 96-well plates containing 100  $\mu\text{l}$  of Mueller-Hinton broth and 5  $\mu\text{l}$  of bacterial suspension. Each test was performed in triplicate and the plates were incubated in an oven at 37°C for 24 hours. As an experimental control, wells with only Mueller-Hinton broth, wells with only essential oils, wells with Mueller-Hinton broth and bacterial suspension, and wells with the bacterial suspension with 100  $\mu\text{g/mL}$  Gentamicin were used. In the wells where inhibition of bacterial growth was observed, that is, in which the culture medium remained non-turbid, 20  $\mu\text{L}$  of the suspensions were collected and seeded on Mueller-Hinton agar plates in an oven at 37°C for 24 hours, for analysis of bacterial growth and CBM. Descriptive statistical analysis was carried out to assist in describing the observed phenomenon. Average concentrations were obtained for the essential oils of oregano, pink pepper, and tea tree, 2500  $\mu\text{g/mL}$ , 625  $\mu\text{g/mL}$ , and 5000  $\mu\text{g/mL}$  against the *E. coli* strains and 1250  $\mu\text{g/mL}$ , 2500  $\mu\text{g/mL}$ , and 625  $\mu\text{g/mL}$  for the gram-positive bacteria *S. aureus*, respectively. Regarding the bactericidal efficiency of oregano essential oil, it was higher on *S. aureus* strains. The results regarding the CBM of pink pepper essential oil indicated values of 625  $\mu\text{g/mL}$  for *E. coli* and 2500  $\mu\text{g/mL}$  for *S. aureus*. As for tea tree essential oil, MBC values of 5000  $\mu\text{g/mL}$  were obtained for *E. coli* and 625  $\mu\text{g/mL}$  for *S. aureus*. Therefore, it is concluded that there is antimicrobial effectiveness of these plant extracts against the bacteria *E. coli* and *S. aureus*.

**Keywords: natural additive, microorganisms, antimicrobial action, E-coli, S-aureus**