EFFECT OF OREGANO ESSENTIAL OIL AND INFUSION ON BACTERIAL BIOFILMS ON STRAINS ISOLATED FROM STOOL OF CHILDREN WITH DIARRHEA.

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Introduction. Biofilms are sessile communities formed by unicellular organisms jointed irreversibly to a substrate and among them. They have been described as conferring multi-resistance to a number of pathogenic bacteria which cause persistent infections (1). Strategies to eradicate biofilms can consist on the inhibition of its development or its disaggregation; the former may attack quorum sensing factors, adhesion, etcetera, and the later requires enzyme activity, because the Exopolysaccharide matrix had already been formed (2). This study aims to detect the effects of Mexican oregano essential oil and oregano aqueous infusion in the formation and disaggregation of biofilms formed by multi-resistant bacteria, which were isolated from children with gastrointestinal infection processes.

Methods. The characterization of four bacterial strains, named 6SS, 13.2SS, 18.2SS and 35mck was done by Gram staining, macroscopic and microscopic morphology and a set of biochemical test for Gram negative bacteria. To test the inhibition in biofilms formation, bacterial strains were inoculated in microplates containing TSB with two different concentrations of oregano infusion and oregano essential oil (concentrations were lower than the minimal bactericidal concentration). Microplates were incubated and after incubation, culture was carefully discarded, and the wells were tested for adhesion of biofilms, by the methylene blue staining procedure. In order to evaluate the effect on already formed biofilms, bacterial strains were grown in 50 ml Falcon tubes, with a glass slide inside, so that the biofilm could be formed. Glass slide was then submerged for 15 minutes in a solution of either Mexican oregano infusion or essential oil; biofilms from slides were then washed into buffer solution and bacterial counts were obtained by serial dilution and spread plate on TSA.

Results. Bacterial strains were identified as *Salmonella spp.*, except for 13.2SS, which was identifies as *Enterobacter spp.* based on biochemical tests. For the biofilm inhibition test, results showed that oregano infusion inhibited biofilm formation at lower and higher concentrations, suggesting a tolerance to infusion components, until the components can actually inhibit bacteria growth and biofilm formation. These results were not found with the essentiall oil, were the effect was concentration-dependent.

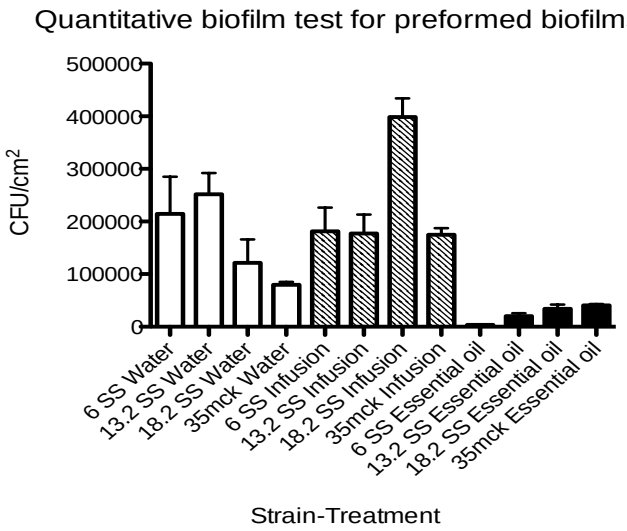

Fig. 1. The effect to infusion, water, and essential oil is shown. A strain shown a significant increment in the disaggregation with infusion.

Figure 1 shows the results of oregano infusion and essential oil on pre-formed biofilm. It is observed a higher bacterial count with the infusion and a significative decrease with the essential oil.

Conclusions. Mexican oregano aqueous infusion inhibited the formation of biofilms in multiple resistant clinical isolates, but when preformed biofilms were exposed to oregano infusion, an increase in bacterial cell was observed as compared to the control. This effect was not observed with oregano essential oil, that was bactericidal and the effect was concentration-dependent.

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