

## Inhibitory effect of extracts of *Croton antisyphilticus* in biofilm production of *Staphylococcus aureus* causing bovine mastitis.

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**Introduction**: The bovine mastitis is the main disease that affects dairy herds. Staphylococcus aureus as the etiologic agent is highly prevalent and difficult to control. Among the various virulence factors of the pathogen the ability to form biofilms, which are bacteria adhered to a surface surrounded by a matrix of organic polymers, gives great protection by preventing the action of phagocytic cells and the direct action of antibiotics on them. Because of the need to discover novel compounds that are effective for the control and treatment of disease, recent studies have demonstrated significant results in the inhibition of reduction of biomass or biofilm using vegetable extracts and isolated compounds. **Objective**: To evaluate the inhibitory potential of extracts of *Croton* antisyphilticus in biofilm production of Staphylococcus aureus isolated from milk of animals diagnosed with bovine mastitis. Methods: The extracts were prepared from the roots and leaves of *C. antisyphiliticus* using the solvent chloroform. Solubilization was added to dimethylsulfoxide (DMSO) and Tween 80. The extracts were tested at a concentration of 5 mg / ml. The gentamicin sulfate (30 mg / ml) was used as control. Ten strains of Staphylococcus aureus genotypically and phenotypically identified as producing biofilm were used, being a standard strain ATCC 25923 (control) also producing biofilm. After standardization of inoculum  $(10^5 \text{ CFU} / \text{mL})$  were placed in 96 well plates containing culture medium Brain Heart Infusion (BHI) containing 2% glucose and the extracts analyzed. Incubation was at 37 ° C for 24 hours under agitation (120rpm). All assays were performed in triplicate, and controls (inoculum, vehicle and plant extracts). For measurement of biomass, the technique of crystal violet was applied, followed by reading spectrophotometer (492 nm). **Results**: Data were calculated as percentage of inhibition in the formation of biofilm, while the inoculum as a reference control (100% of biofilm formation). According to the average obtained from the chloroform extract of root and leaf C.antisyphiliticus (5 mg / mL) inhibited respectively 87.59% and 88.04% compared to control while gentamicin inhibited 87.02 % biofilm formation. Conclusion: The results show that the extracts evaluated have considerable ability to reduce the biomass formed by S. aureus under the conditions tested, surpassing the average presented by the antibiotic in question, a concentration six times lower. Thus, considering the problems involved in this important bacterial virulence factor, more detailed studies are needed to in order to identify substances of plant origin capable of acting effectively on microorganisms, minimizing the use of overt antibiotics in animal production.

Keywords: Croton, Staphylococcus aureus, biofilm.

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