

TITLE: USE OF EXTRACT *Hypericum brasiliense* ON THE FORMATION OF BIOFILMS BY SUBSPECIES OF *Staphylococcus schleiferi* ISOLATED OF CANINE OTITIS IN THE STATE OF RIO DE JANEIRO.

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ABSTRACT:

Staphylococcus can be commonly found inhabiting the microbiota of the skin and mucosa of the upper respiratory tract of mammals and birds. Occasionally, staphylococci can cause serious infections in their hosts. In dogs, *Staphylococcus schleiferi* is one of the most important species, causing topical infections such as pyoderma and otitis. *S. schleiferi* can be separated into two subspecies: *S. schleiferi* subsp. *coagulans* and *S. schleiferi* subsp. *schleiferi*. A characteristic species of this genus are its virulence factors. Biofilm formation leads to difficult-to-treat infectious diseases (due to biofilm protection against antimicrobials and host defenses). The presence of resistant strains in dogs generates great problems, such as: limited treatment options in veterinary medicine and the fact that these species serve as reservoir for resistance genes and represent a risk of zoonotic transmission. The genus *Hypericum* has several species with proven antibiotic activity, such as *H. brasiliense*, which has been shown to be promising in previous studies. This study aimed to evaluate the activity of *H. brasiliense* in combating biofilm formation of *S. schleiferi* subspecies isolated from dogs affected by pyoderma or otitis. A collection of 9 strains of *S. schleiferi*, including five of *S. schleiferi* subsp. *schleiferi* and four of *S. schleiferi* subsp. *coagulans*, were first seeded in TSA. After incubation at 37°C (24 hours), Minimum Inhibitory Concentration Determination (MIC) assays were carried out by microdilution in broth. The MIC is the lowest concentration of the derivative that inhibits visible growth of the bacterial culture. The reading was visual by Resazurin dye 0.01%, where the color pink indicates bacterial growth and blue, absence of growth. The extract concentration was 512 µg/mL at 4 µg/mL. Subsequently, the minimum bactericidal concentration (MBC) test was performed, defined as the lowest concentration capable of eliminating 99.9% of the inoculum. The extract obtained MIC results for *S. schleiferi* subsp. *schleiferi* strains, between 32 and 64 µg/mL, and MBC between 32 and 128 µg/mL. About *S. schleiferi* subsp. *coagulans*, the extract obtained MIC results between 16 and 64 µg/mL and MBC between 32 and 128 µg/mL. The *in vitro* antibiofilm activity of the extract against the same strains was evaluated by the protocol of SILVA et al. (2010) with adaptation. The results with the extract of *H. brasiliense* suggest good activity in the fight against biofilm formation. The extract obtained a better result in the concentration of ½ MIC (32 µg/mL) in *S. schleiferi* strains, besides being better than the results found for the control of antibiotics (vancomycin). In conclusion, the extract of *H. brasiliense* has action in the fight against the formation of biofilms of subspecies of *S. schleiferi*. It is an interesting study product to combat infections caused by biofilm-forming bacteria, and may be an alternative for multiresistant strains.

Keywords: biofilm, *Hypericum brasiliense*, extract activity.

Development Agency: Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – CAPES.