TITLE: ACTIVITY OF BIOGENIC SILVER NANOPARTICLES AGAINST ISOLATES OF *PROTOTHECA BOVIS*

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ABSTRACT:

Algae of the Prototheca genus has been highlighted as a causative agent of infections in animals and humans, demonstrating the importance of this pathogen in One Health. The main form of presentation in production animals is bovine mastitis, caused mostly by Prototheca bovis and Prototheca blaschkeae. The economic losses in livestock caused by this agent have been increasing due to the greater number of infections combined with the lack of effective treatment. Infections in these animals are not responsive to antimicrobials, and the recommended protocol for the control of protothecosis in herds is chemical drying of the affected quarter and/or the disposal of the animals. Thus, the search for alternatives for the treatment of infections by *Prototheca* spp. is necessary. Silver nanoparticles has been widely tested due to antimicrobial effects against various microorganisms. Therefore, this study aimed to investigate the inhibitory and algaecide effects of a biogenic silver nanoparticle (Bio-NPAg) against P. bovis isolates of bovine mastitis. The microplate minimum concentration method was used for testing Bio-NPAg (60 µg to 1.875 µg mL⁻¹) against *P. bovis* isolates(n=20)(10⁶ CFU mL⁻¹) ¹/well). Microplates were incubated at 37 °C/24 h. After, an aliquot from each well was transferred to a BHI agar plate to assess the growth viability of Prototheca sp. Minimum algicidal concentration (MAC) was defined as the lowest concentration of Bio-NPAg that prevented algae growth on BHI agar at 37°C/48 h. Minimum inhibitory concentration (MIC) was defined as the lowest concentration of Bio-NPAg in which there was no metabolic activity of Prototheca sp., determined by the absence of color change of the resazurin reagent (30µl/24 h). In only 5% of the tested isolates (1/20) it was not possible to determine the MIC and MAC (> 60 μ g mL⁻¹ of Bio-NPAg). In 20% (4/20) of the tested P. bovis isolates, the MIC was $60\mu g \text{ mL}^{-1}$, in 50% (10/20) the MIC was equal to 30 μ g mL⁻¹ and in 25% (5/20) the MIC was 15 μ g mL⁻¹. In 30% (6/20) of the P. bovis isolates tested, the MAC was 60 µg mL⁻¹, in 45% (9/20) the MAC was equal to 30 µg mL⁻¹, in 20% (4/20) the MAC was 15 μ g mL⁻¹. The preliminary results demonstrate that Bio-NPAg has potential for the control of infections by Prototheca sp., requiring further studies to confirm these results.

Keywords: biogenic silver nanoparticles, protothecosis, algae, mastitis

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