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DISINFECTANT FORMULATION WITH ANTIBIOFILM ACTIVITY BASED ON SILVER NANOPARTICLES SYNTHETIZED IN SITU WITH AQUEOUS EXTRACTS OF Minthostachys mollis Griseb.

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Currently, the disinfectants used in hospital environments do not ensure the complete elimination of bacteria and/or biofilms on various biomedical products (such as probes, endoscopes, catheters, etc.). Microbial resistance has increased worldwide in recent years. Therefore, our work focuses on developing new eco-friendly disinfectant formulations based on silver nanoparticles (AgNP) combined with plant extracts from native flora. The aerial parts of Minthostachys mollis Griseb. ("Peperina," Lamiaceae) were collected during the flowering season from the Los Quebrachillos Cabana Reserve in Unquillo, Córdoba Province, Argentina. The plant material was dried in the shade, ground into a powder, extracted with distilled water (by maceration), and then filtered by gravity. Green synthesis of the nanoparticles was performed using 50 ?L of the aqueous plant extract and 150 ?L of a 10 mM AqNO? solution at 85°C for 10 minutes. The antimicrobial activity of Mm@AgNP and the aqueous extract of Minthostachys mollis evaluated against the reference was Staphylococcus aureus ATCC 29213, methicillin-resistant Staphylococcus aureus ATCC 43300 (MRSA), Escherichia coli ATCC 25922, ESBL-producing Escherichia coli ATCC 35218, and a clinical multidrug-resistant strain of Escherichia coli, with an inoculum of 10? CFU/mL in phosphate-buffered saline (PBS). After the treatments, colonies were counted on tryptone soy agar plates following 24 hours of incubation at 37°C. The antimicrobial capacity of Mm@AgNP was also evaluated in planktonic culture and in biofilms of a clinical strain of S. aureus MR 9455. Controls without nanoparticles were also run in parallel. We demonstrated that in planktonic cells, complete inhibition was achieved within 5 minutes of treatment with Mm@AgNP across all the studied strains. The inhibition time was reduced to 2 minutes when Mm@AqNP suspensions were 10-fold concentrated compared to the control. The extract alone did not show antibacterial activity at the evaluated time. A mature biofilm exposed to a 10-fold concentration of Mm@AqNP suspension for 24 hours revealed damage to the matrix integrity and a reduction in the density of cell groups. The morphology of the cocci remained intact in the controls, which showed a matrix density markedly higher than in the treated samples. The high

activity detected within minutes of incubating the bacterial suspension with the nanoparticles is promising and suggests their potential as a disinfectant. Thanks to Dra. María Claudia Luján from IMBIV-CONICET for the collection and identification of the plant material, SECyT-UNC and CONICET for financial support and LAMARX (FAMAF - UNC) for SEM analysis.

Palabras clave: green synthesis - silver nanoparticles - antimicrobial activity – eco-friendly disinfectants