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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 µL of the aqueous plant extract and 150 µL of a 10 mM AgNO<sub>3</sub> solution at 85°C for 10 minutes. The antimicrobial activity of Mm@AgNP and the aqueous extract of *Minthostachys mollis* was evaluated against the reference strains *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<sup>7</sup> 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@AgNP 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@AgNP 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