# Ciprofloxacin- and gentamicin-mediated inhibition of Pseudomonas aeruginosa biofilms is enhanced when combined the volatile oil from Eucalyptus camaldulensis

Al-Moghira K. Al-Qaysi, 1 Mushtak T.S. Al-Ouqaili, 2 Safaa A.L. Al-Meani3 1Department of Biology, College of Education for Pure Sciences, University of Anbar Ramadi, Iraq 2Department of Microbiology, College of Medicine, University of Anbar, Ramadi, Iraq 3Department of Biotechnology, College of Science, University of Anbar, Ramadi, Iraq Corresponding author: Mushtak T.S. Al-Ouqaili ph.dr.mushtak_72@uoanbar.edu.iq ABSTRACT Pseudomonas biofilms cause therapeutic failures in many clinical infections. This study investigated the combined actions of the volatile oil from Eucalyptus camaldulensis and antibiotics (ciprofloxacin and gentamicin) against planktonic and biofilm populations of Pseudomonas aeruginosa. Further, the quorum sensing genes of P. aeruginosa, grown as biofilms, were also investigated. The minimum inhibitory concentrations (MICs) and biofilm inhibitory concentrations (BICs) of gentamicin, ciprofloxacin, and/or the volatile oil of Eucalyptus camaldulensis were determined against planktonic and biofilm populations of clinical P. aeruginosa isolates. Further, the fractional inhibitory concentrations (FICs) were determined for the antibiotics and volatile oil using a modified checkerboard assay. The bacterial genomic DNA was extracted from biofilm-producing

isolates and the target quorum sensing genes were amplified using polymerase chain reaction. Out of 96 biofilm-producing isolates, 43 (44.8%), 29 (30.2%), and 24 (25%) were, respectively, strong, intermediate, and weak biofilm producers. The ciprofloxacin BICs were 30–200-fold higher than the ciprofloxacin MICs. Ciprofloxacin and the volatile oil demonstrated a synergistic effect against both planktonic and biofilm populations. Although the volatile oil and gentamicin also demonstrated a synergistic effect against planktonic cells, only an additive effect was observed against biofilm cells. There was a significant relationship between the frequency of quorum sensing genes, rhlLR and lasLR, and the MICs of piperacillin, with P values of 0.043 and 0.032, respectively. The ciprofloxacin MIC was significantly correlated with the presence of lasLR (P = 0.031) and the ceftazidime MIC was correlated with the presence of lasLR (P = 0.023). The volatile oil from E. camaldulensis leaves has antibacterial effects on P. aeruginosa biofilms, alone and in combination with ciprofloxacin. There is a high correlation between the presence of quorum sensing genes (LaslR and rhlLR) and biofilm production, and with pipracillin resistance.

Keywords: