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BIOSURFACTANTS FROM *Pseudomonas aeruginosa* MM OBTAINED FROM FRYING OIL: USE IN SURFACTANT-ENHANCED BIOREMEDIATION

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Pseudomonas aeruginosa MM is a hydrocarbon-degrading, surfactant-producing bacterium isolated from an urban stream in Moreno, Buenos Aires Province. Previous studies demonstrated that *P. aeruginosa* MM could produce both rhamnolipids and lipopeptides using raw sunflower oil (RSO) as a carbon source. When a crude extract (SCE) of these biosurfactants was used in surfactant-enhanced bioremediation (SER), 47% of hydrocarbons were removed compared to an untreated control. Given that the high production costs of bacterial biosurfactants limit their use in SER, this study aimed to obtain a surfactant crude extract (SCE) using an inexpensive carbon source, such as frying oil. *P. aeruginosa* MM was cultured in E2 minimal medium supplemented with sterile frying oil as a carbon source. Cultures were incubated at 37°C for 2 days at 150 rpm. After incubation, a cell-free supernatant was obtained by centrifugation (10 min at 12,000 rpm). The cell-free supernatant was acidified, incubated at 4°C overnight, and then centrifuged at 12,000 rpm for 20 min. The resulting pellet was resuspended in 0.1M Tris-HCl, pH 8, and extracted thrice with 1 volume of ethyl acetate. The solvent was evaporated, and the remaining dry compounds were resuspended in distilled water to obtain the frying oil crude extract surfactants (F-SCE). The critical micellar concentration (CMC) of the F-SCE was measured using a Du Nouy tensiometer, yielding a CMC of 137 µg/mL showing better performance than the obtained with the RSO-SCE (CMC of 317 µg/mL). About the surface tension (ST) obtained at the CMC, the F-SCE reached a ST of 38.5 mN/m while the RSO-SCE was 33.5 mN/m. Finally, the F-SCE was tested as an additive in SER microcosm assays. For these tests, 10 g of soil was supplemented with KNO₃ and K₂HPO₄, adjusted to 60% field capacity, and artificially contaminated with 10% v/w diesel. Two sets of five units each were designed: one without surfactants (control) and one with F-SCE at a concentration of twice the CMC relative to the water present in the microcosm. The microcosms were incubated for 24 days at 24°C. After incubation, the remaining diesel was extracted and analyzed by GC-FID. The results showed a diesel degradation of 59.2 ± 6.6% compared to the control without surfactant. This study demonstrated that using frying oil as a carbon source for *P. aeruginosa* MM biosurfactant production improved the characteristics of the SCE

derived from raw sunflower oil and reduced production costs.

Palabras clave: Pseudomonas - Biosurfactants - Bioremediation - Hydrocarbon