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A non-toxic microbial surfactant from *Marinobacter hydrocarbonoclasticus* SdK644 for crude oil solubilization enhancement

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ABSTRACT

This study aims to investigate the ability of a biosurfactant produced by *Marinobacter hydrocarbonoclasticus* strain SdK644 isolated from hydrocarbon contaminated sediment to enhance the solubilization rate of crude oil contaminated seawater. Phylogenetic analysis shows that strain SdK644 was very closely related to *M. hydrocarbonoclasticus* with 16S rRNA gene sequence similarity of 97.44%. Using waste frying oil as inducer carbon source, the producing biosurfactant by strain SdK644 was applied to improve crude oil solubilization in seawater. The preliminary characterization of the produced biosurfactant by FT-IR analysis indicates its possible classification in a glycolipids group. Results from crude oil solubilization assay showed that SdK644 strain biosurfactant was 2-fold greater than Tween 80 surfactant in crude oil solubilization and 12-fold higher than seawater control, as shown by GC-MS analysis of aliphatic compounds. Furthermore, this bioactive compound was shown to be nontoxic against *Artemia* larvae in short-term acute toxicity bioassay. Generally, the results showed the possible use of *M. hydrocarbonoclasticus* strain SdK644 biosurfactant in bioremediation processes of the marine environments.

1. Introduction

Petroleum pollution of the environment is of grave risk because petroleum hydrocarbons are toxic to all forms of life. The contamination of the environment by crude oil is quite common because of its widespread use and its accompanying disposal operations and accidental spills (Zahed et al., 2010). The total input of petroleum hydrocarbons into the oceans from all sources is about 1.300.000 t per year. Alone, natural seeps account for 46% and 37% by all activities associated with consumption of petroleum products. Adding to all this, during transportation of petroleum products, accidental spills and operational discharges of cargo oil contributes with 12% of the total flux discharged, followed by far by extraction processes (3%) (NRC, 2003).

Crude oil -a heterogeneous mixture of hydrocarbons- consists mainly of alkanes, cycloalkanes, and aromatics. Low amounts of resins (nitrogen, sulfur, and oxygen compounds), and asphaltic fraction (partially oxygenated and highly condensed) exist also in crude oil with varying rates depending on the nature of the oil, light or heavy (Tyagi et al., 2011; Weng et al., 2015). Microorganisms capable of degrading hydrocarbons have a ubiquitous existence (NRC, 2003; Vandecasteele, 2005; McGenity et al., 2012). The principal-hydrocarbon degrader bacteria in marine environments are: *Alcanivorax*, *Marinobacter*, *Thallassolituus*, *Cycloclasticus*, *Oleispira,* and a few others (Yakimov et al., 2007; Acosta-González and Marqués, 2016).

The biodegradation kinetics of crude oil in seawater is controlled by numerous factors such as crude oil composition and concentration, temperature, oxygen, nutrients supply, salinity, pH, and oil availability to microorganisms (Vandecasteele, 2005). Petroleum hydrocarbons have a limited bioavailability because they are mostly insoluble in water (Chen et al., 2013). To overcome this kinetic limitation, the application of synthetic surfactants or biosurfactants can promote this availability by reducing interfacial tensions between the two immiscible phases, which leads to increase the surface area of oil slick and therefore improve the solubility of hydrocarbons (Urum and Pekdemir,

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