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### Enhanced oil recovery under laboratory conditions using biosurfactant-producing microorganisms

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Oil recovery comprises a primary phase, which produces oil using the natural pressure drive of the reservoir, and a secondary phase, which includes the injection of water to improve the flow of oil to the wellhead [1,2]. While primary recovery produces 5-10% of the original oil in place, recovery efficiencies in the secondary stage vary from 10% to 40% [1]. Most of the unrecovered oil (up to two-thirds of the total oil reserves) is trapped in the reservoir pores by high capillary forces [2]. Microbial Enhanced Oil Recovery (MEOR) is a tertiary oil recovery process where microorganisms and their metabolites are used to retrieve unrecoverable oil from mature reservoirs. Stimulation of biosurfactant production by indigenous or injected microorganisms can reduce the capillary forces that retain the oil into the reservoir. In this work, a sand pack column model was designed to simulate the oil recovery operations in oil reservoirs and evaluate the mobilization of residual oil by microorganisms. Three *Bacillus subtilis* strains (309, 311 and 573), previously isolated from crude oil samples, were used in this study. They grow and produce extracellular biosurfactants at 40°C under anaerobic conditions in medium supplemented with hydrocarbons. Biosurfactants produced by those isolates reduce the surface tension of water from 72 to 30 mN/m, exhibit emulsifying activity and are not affected by exposure to high temperatures (121°C) which makes them good candidates for application in biosurfactant mediated MEOR. Acrylic columns (250 ml) packed with acid washed sand were first flooded with water, after that saturated with paraffin, and then washed with water to remove the excess of paraffin. Afterwards, the isolates were injected into the columns with the optimized medium and incubated at 40°C. After 14 days, the columns were flooded with water and the additional oil recovery (AOR) was calculated as the percentage of paraffin recovered. AOR using *B. subtilis* 309, 311 and 573 was  $35.0 \pm 1.0$  %,  $23.5 \pm 1.2$  % and  $19.8 \pm 1.9$  %, respectively. The results obtained suggest that stimulation of biosurfactant production by these strains in the oil reservoir can contribute to mobilize entrapped oil.

[1] Sen R. 2008. *Progress in Energy and Combustion Science* 34: 714-724.[2] Brown LR. 2010.. *Current Opinion in Microbiology* 13: 1-5.