



INDIAN INSTITUTE OF TECHNOLOGY GUWAHATI  
SHORT ABSTRACT OF THESIS

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Programme of Study : **Ph.D.**  
Thesis Title: **Exploring inherent bacterial strains for potential application in biosurfactant production and crude oil biodegradation**  
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Thesis Submitted to the Department/ Center : **Department of Biosciences and Bioengineering**  
Date of completion of Thesis Viva-Voce Exam : **02-09-2022**  
Keywords for description of Thesis Work : Crude oil bioremediation, Biosurfactant, Biosorption, Surface modification, Alkane hydroxylase, Catechol dehydrogenase, Degradation kinetics

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The universal and pervasive need for oil as an energy source has caused excessive exploitation, escorted by tragic accidental spills and chronic contamination of the marine ecosystem. Biodegradation is an eco-friendly and effective tool for the remediation of spilled oil; however, the slower degradation rate limits its exploitation for practical purposes. The selection of potential biodegraders is the key to addressing this challenge. This thesis aimed to explore such crude oil-degrading bacteria inherent to oil-contaminated sites and improve their biodegradation efficacy by optimizing the culture conditions and nutrient requirements for maximum biosurfactant production. In this direction, the biosurfactant production ability of an indigenous bacterial strain *Agrobacterium fabrum* SLAJ 731, was explored. Optimization of bacterial biosurfactant production ability was achieved using the One factor at a time approach (OFAT), where pH 6, 37°C using Glucose and Yeast extract in 2:1 (w/v) ratio were obtained as optimal culture requirements for maximum biosurfactant production of 5.77 g/L. Studies revealed a synergistic effect of biosurfactant production on bacterial crude oil biodegradation efficiency. Thus to unveil the role of biosurfactants in crude oil bioremediation, another isolated oil-degrading strain, *Bacillus subtilis* RSL 2, was preliminarily optimized for the maximum biosurfactant production using the Response surface methodology- central composite design (RSM-CCD) technique. A high 3.5 g/L biosurfactant concentration was optimized at pH 4, 25°C using Crude oil (1 g/L) culture conditions. The produced biosurfactant was investigated for its effect on oil biodegradation in two modes (a) prior addition to media followed by microbial inoculation (sequential mode) and (b) simultaneous addition with inoculum. The findings revealed that biosurfactants (<CMC) improved oil mobilization, making it more bioavailable and enhancing oil biodegradation to 72 % when added simultaneously with inoculum.

Further, a microbial consortium was designed using *A. fabrum* SLAJ 731, *B. subtilis* RSL2, and an exogenous oil-degrader *Pseudomonas aeruginosa* MTCCP7815. The biodegradation kinetics of single aliphatic (Hexadecane), aromatic (Phenanthrene) and the binary mixture as co-contaminants by axenic cultures of *A. fabrum* SLAJ 731, *B. subtilis* RSL2 and *P. aeruginosa* P7815 and their consortium were explored. An integrated kinetic model combining first-order exponential decay and the Monod equation was well fitted to the biodegradation results. The maximum degradations (> 90%) of both substrates were observed for the microcosm compared to axenic cultures, indicating the selected strains' synergistic effects. Immobilization techniques using non-toxic, economical, and eco-friendly sugarcane bagasse were performed to strengthen the crude oil biodegradation efficiency further. The surface hydrophobicity of the bagasse was improved by forming octyl self-assembled monolayers to enhance the surface-oil interactions (adsorption). Henceforth, an integrated biodegradation strategy using a faster removal method, adsorption, was addressed to overcome the slower bacterial biodegradation rate. The biodegradation results fitted well to combined exponential decay and Monod's degradation model, inferencing a simultaneous biosorption and biodegradation phenomenon. The integrated approach synergistically improved overall crude oil remediation, which correlated with microbial degradative enzyme activity and biosurfactant production. Hence, hydrophobic biosorbents immobilized potential oil-degrading and biosurfactant producing consortium shall be used for bioremediation of oil-contaminated sites in the foreseeable future.