

Mohammad A. Zahed<sup>1</sup>  
Hamidi A. Aziz<sup>1</sup>  
Mohamed H. Isa<sup>2</sup>  
Leila Mohajeri<sup>1</sup>

<sup>1</sup>School of Civil Engineering, Universiti Sains Malaysia, Nibong Tebal, Penang, Malaysia

<sup>2</sup>Civil Engineering Department, Universiti Teknologi PETRONAS, Tronoh, Perak, Malaysia

## Research Article

# Response Surface Analysis to Improve Dispersed Crude Oil Biodegradation

In this research, the bioremediation of dispersed crude oil, based on the amount of nitrogen and phosphorus supplementation in the closed system, was optimized by the application of response surface methodology and central composite design. Correlation analysis of the mathematical-regression model demonstrated that a quadratic polynomial model could be used to optimize the hydrocarbon bioremediation ( $R^2 = 0.9256$ ). Statistical significance was checked by analysis of variance and residual analysis. Natural attenuation was removed by 22.1% of crude oil in 28 days. The highest removal on un-optimized condition of 68.1% were observed by using nitrogen of 20.00 mg/L and phosphorus of 2.00 mg/L in 28 days while optimization process exhibited a crude oil removal of 69.5% via nitrogen of 16.05 mg/L and phosphorus 1.34 mg/L in 27 days therefore optimization can improve biodegradation in shorter time with less nutrient consumption.

**Keywords:** Bioremediation; Hydrocarbon; Oil spill; Petroleum; Total petroleum hydrocarbons (TPH)

*Received:* October 2, 2009; *revised:* August 24, 2010; *accepted:* January 18, 2011

**DOI:** 10.1002/clen.200900215

## 1 Introduction

Biodegradation of petroleum hydrocarbons is one of the most important processes involved in weathering and the eventual removal of petroleum from the environment, particularly for the non-volatile components of petroleum. A number of scientific articles have covered various aspects of this process and the environmental factors that influence the rate of biodegradation [1–3]. Petroleum biodegradation is carried out by microorganisms capable of utilizing hydrocarbons as a source of energy and carbon. These microorganisms are ubiquitous in nature and are capable of degrading various types of hydrocarbons: Short-chain, long-chain and numerous aromatic compounds, including polycyclic aromatic hydrocarbons. Bioremediation has been defined as “the act of adding materials to contaminated environments to cause an acceleration of the natural biodegradation processes” [4]. The use of bioremediation for large-scale field application gained significant attention as beaches contaminated with oil from the Exxon Valdez spill, Prince William Sound, Alaska in 1989, were seeded with fertilizer to promote the growth of hydrocarbon-degrading bacteria [5].

One factor which affects the environmental cycling processes of petroleum is the addition of dispersant chemicals. In marine ecosystems, nutrient limitation is generally correlated with low background levels of nitrogen and phosphorus in seawater [6]. Oil dispersants are mixtures of surfactants and solvents which effec-

tively disseminate oil in the water column, creating small oil droplets [7, 8]. Treatment of oil spills with dispersants in temperate marine environments has been common practice for many years. A major reason for using these chemicals is to prevent spilled oil from reaching the shore. However, it is thought that dispersed oil presents an increased toxicity to marine life compared to untreated oil, as a result of the detrimental effects of surfactants and elevated hydrocarbon dissolution [9]. Thus, benthic and pelagic organisms may also be exposed to the harmful impacts of both oil and dispersant [10–12]. Lindstrom and Braddock [13], however, reported that nutrient addition may be important to consider in environmental application of the Corexit 9500 dispersant, since the dispersant appears to be a good microbial substrate.

Response surface methodology (RSM) is a useful mathematical and statistical method for analyzing the effects of several independent variables on process outcomes (response) [14, 15]. In many processes, the relationship between the response and independent variables is usually unknown; therefore, the first step in RSM is to approximate the function (response) in terms of independent variables. Usually, this process employs a low-order polynomial equation in a pre-determined region of the independent variables, which will later be analyzed to identify the optimum values of independent variables for the best response.

Recently, RSM has been used in bioremediation optimization studies such as optimizing biodegradation of weathered crude oil in coastal sediments [16], Enhancement biodegradation of *n*-alkanes from crude oil contaminated seawater [17], optimization of nutrient component for diesel oil degradation [18] phenol degradation by *Pseudomonas putida* [19], and bioremediation of a soil contaminated by lindane [20]. The present study employed RSM for analyzing the effects of nutrient supplementation, dispersant addition and degradation time on biodegradation of dispersed crude oil (DCO).

**Correspondence:** Professor H. A. Aziz, School of Civil Engineering, Universiti Sains Malaysia, 14300 Nibong Tebal, Penang, Malaysia  
E-mail: cehamidi@eng.usm.my

**Abbreviations:** DCO, dispersed crude oil; RSM, response surface methodology; TPH, total petroleum hydrocarbons