Iranian Journal of Chemical Engineering
Vol. 9, No. 4 (Autumn), 2012, IAChE

Study of Enhanced Bioremediation in Treatment of Gas Condensates Contaminated Soil

J. Shayegan[*], *A. Babaee*
School of Chemical and Petroleum Engineering, Sharif University of Technology, Tehran, Iran

Abstract

In recent decades, large amounts of hydrocarbon derivations have contaminated the environment due to industrial developments and neglecting the environmental issues. In this study, the ability of biological removal of hydrocarbon pollution from contaminated soil around Sarkhun gas refinery was investigated. This study was done for seven samples with three different nutrient ratios and by moisture controlling and continuous aeration. During experiments, concentration of Total Petroleum Hydrocarbon (TPH) was measured. Results showed that with a nutrient ratio of 100:5:1 for C:N:P during an 18-day remediation period, the mean contaminant removal was about 50%. By using these results a practical plan was suggested for running in real scale situations. Developing a model was done by considering the Monod model as microbial growth model. Results showed good accordance to empirical data.

Keywords: *Enhanced Bioremediation, Contaminated Soil, TPH, Gas Condensates, Sarkhun Gas Refinery, Monod model*

1. Introduction

Hydrocarbon contamination results from leakage of aboveground and underground storage tanks, spillage during transport of petroleum products, petroleum and gas refineries, and other accidental releases. Petroleum contains hazardous chemicals such as benzene, toluene, ethylbenzene, xylenes, and naphthalene that can be hazardous to the health of plants, animals, and humans [1-4].

Petroleum-contaminated soil is currently being treated using physical, chemical, and biological processes. Most of the physical methods currently used for treatment of contaminated soils are expensive [3]. Chemical treatment includes direct injection of oxidizing agents into contaminated soil and groundwater [5], thereby altering native aquatic chemistry [5]. So, the use of microbial potential for the remediation of toxic compounds from soil (bioremediation) is now accepted as an alternative to conventional methods [6].

Bioremediation is a process in which the microorganisms consume organic

* Corresponding author: shayegan@sharif.edu

contaminants and convert them to innocuous end products. The activity of naturally occurring microbes is stimulated by circulating water-based solutions through contaminated soils. This process enhances in-situ biological degradation of organic contaminants or immobilization of inorganic contaminants. Nutrients, oxygen, or other amendments may be used to enhance bioremediation and contaminant desorption from subsurface materials [7].

Bioremediation of contaminants can be accomplished by two methods: bioaugmentation and biostimulation. The process of bioaugmentation, as it applies to remediation of petroleum hydrocarbon contaminated soil, involves the introduction of microorganisms that have been cultured to degrade various chains of hydrocarbons into a contaminated system. The cultures may be derived from the contaminated soil or they may be obtained from a stock of microbes that have been previously proven to degrade hydrocarbons. Once introduced into the system, the cultured microorganisms selectively consume the hydrocarbons. The process of biostimulation introduces additional nutrients in the form of organic and/or inorganic fertilizers into a contaminated system which increases the population of the indigenous microorganisms [8].

Bioremediation of petroleum-contaminated soils has been investigated since the late 1940s but interest in the field did not become widespread until the Exxon Valdez oil spill in 1989 [9 and 10]. Consequently, there have been a large number of studies conducted and bioremediation has almost always been found to be an effective treatment of hydrocarbon-contaminated sites [1, 2, 11 and 12]. In the field of biostimulation, nutrient supplementation for hydrocarbon degradation has traditionally focused on addition of N and P, either organically or inorganically. Because carbon (C) is a major constituent of petroleum fuels, its traditional role in bioremediation research has typically been an index to determine the amount of N and P that needs to be added to reach the optimal C:N:P ratio [5]. Several studies have reported positive effects of biostimulation by nutrient amendment on oil decontamination [13]. However, an understanding of nutrient effects at a specific site is essential for successful bioremediation [13]. Biodegradation rates were reported to depend mainly on the concentration of nitrogenous nutrients in the sediment pore waters, the oil loading and the extent to which natural biodegradation had already taken place [14]. Xu et al., in 2010 compared the efficiency of various biostimulation and bioaugmentation strategies in reducing soil petroleum contamination under laboratory conditions. In his study, inorganic nutrients such as $(NH4)_2SO_4$ and K_2HPO_4 were added to the treatment, 27% of TPH, to give a final C:N:P ratio of 100:10:1 [15]. Previous research has shown that nutrient additions enhance the microbial activity, which causes an increase in the bioremediation rate. However, optimal nutrients stimulate the microorganisms. The objective of this study was to identify the effect of nutrient ratio on the extent of enhanced hydrocarbon biodegradation and develop a suitable model to predict the growth rate of microorganisms and removal rate of total petroleum hydrocarbon from the contaminated soil.

2. Materials and methods

2-1. Soil samples

Sarkhun gas refinery is located at about 50km north-east of Bandar Abbas and refines 15 million STD cubic meters of natural gas per day. For the last 20 years, the surrounding soils of the refinery have been gradually contaminated by gas condensates leaked from pipes, burn pits area and wastewater collection network.

It was concluded that after more than 20 years, environmental adaptation has resulted in growing bioremediation microorganisms within the soil. Seven samples from various highly contaminated locations around the refinery were picked up.

2-2. Experimental design and treatment methods

Three different boxes with a capacity of 1290 cm^3 were used to run the experiments. The dimensions and the function of each box are shown in Fig. 1. For each experiment, 1 kg of contaminated soil and nutrient solutions were added to each box. Knowing the initial TPH (X) as mg C/kg soil and considering the C:N:P ratio (C moles of carbon, N moles of nitrogen, and P moles of phosphorous as main nutrients), the amount of nutrients needed can be calculated. NH_4NO_3 and KH_2PO_4 were used as nitrogen and phosphorous sources.

Using a simple test, the required amount of makeup water for recovering moisture lost due to evaporation was calculated for moisture control. The makeup water was added on an everyday basis in the treatment period. Aeration was done by mixing the soil everyday. The TPH was measured every three days. TPH measurements were based on solvent extraction and absorption against IR ray in a 2940 cm-1 frequency which is taken from EPA41302 and ASTM D3921 standards, and processed with a TPH/TOG Analyzer (Infracal) [16,17].

For further study on biodegradation capability of microorganisms in samples, contaminant removal percent (R) was studied and an empirical model was obtained, as follows:

$$R(\%) = \frac{A_1 \times time(\text{day}) + A_2}{time(\text{day}) + B} \quad (1)$$

In which A_1 is final removal efficiency, A_2 is a factor presenting the initial removal percent (that is zero in all samples here) and B represents the Nitrogen to Phosphorous ratio which is the most important parameter in the above equation.

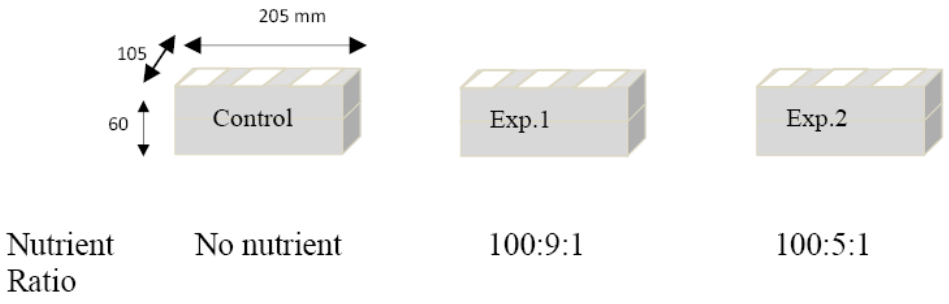

Figure 1. Specifications of boxes used in treatment

2-3. Model description

Model Developing stages were as follows

1. Representing kinetic model of microbial growth and obtaining parameters needed.
2. Developing bioremediation and pollution degradation model.

For these purposes, the following assumptions were made:

- The biological process is aerobic.
- Available oxygen and nutrients needed are constant.
- Temperature and Humidity are constant.
- Pollution is uniformly spread in soil.
- Growth of microorganism follows Monod model.
- The only limited material for biological reactions is organic carbon from contamination.

2-3-1. Kinetic model of microbial activity

Rate of microorganism growth can be represented by equation (2).

$$R_g = \frac{dX}{dt} = \mu X \tag{2}$$

Where R_g is the rate of biomass growth (mass per volume per time), X is the biomass concentration (mass per volume), and μ is the specific growth rate (time^{-1}). μ can be defined by the following equation:

$$\mu = \frac{dX/dt}{X} = \frac{\text{growth rate}}{\text{unit of biomass}} = \frac{\text{mass of new cells synthesized}}{\text{mass of cells present in the reactor} \times \text{time}} \approx \frac{1}{X}\frac{\Delta X}{\Delta t} \tag{3}$$

Monod model that was chosen for microbial growth is:

$$\mu = \mu_{\max}\frac{S}{K_s + S} \tag{4}$$

Where $\mu_{\max}$ is the maximum specific growth rate (time^{-1}), S is the limited substrate concentration (mass/volume), and K_s is the half rate constant or the substrate concentration at a specific growth rate which equals to half $\mu_{\max}$ (mass/volume). Therefore

$$R_g = \mu_{\max}\frac{S}{K_s + S}X \tag{5}$$

Microbial indigenous Decay rate can be defined by equation (6)

$$R_d = -K_d X \tag{6}$$

Where K_d is the indigenous decay coefficient (time^{-1}). Hence, the net rate of biomass growth is

$$R_{\text{net}} = \mu_{\max}\frac{S}{K_s + S}X - K_d X \tag{7}$$

This equation can be written as

$$R_{\text{net}} = Y\frac{KS}{K_s + S}X - K_d X \tag{8}$$

In which Y is the yield coefficient and K is the maximum substrate utilization rate coefficient (time^{-1}).

$$Y = \frac{\text{bacterial growth rate}}{\text{substrate utilization rate}} = \frac{R_g}{R_{su}} \tag{9}$$

$$K = \frac{\mu_{\max}}{Y} \tag{10}$$

Therefore

$$R_{su} = -\frac{KS}{K_s + S} X \tag{11}$$

in which R_{su} is substrate utilization rate (mass per volume per time).

2-3-2. Calculation of biokinetic parameters

To calculate Biokinetic parameters, equation (11) should be rearranged as follows

$$R_{su} = \frac{\Delta S}{\Delta t} = \frac{KS}{K_s + S} X \Rightarrow \left(\frac{X\Delta t}{\Delta S}\right) = \left(\frac{K_s}{K}\right)\frac{1}{S} + \frac{1}{K} \tag{12}$$

Therefore, it is possible to obtain K and K_s by plotting ($X\Delta t/\Delta S$) vs. ($1/S$). Also, the net rate of microbial growth (equation (8)) should be rearranged as follows

$$\left.\begin{aligned} &\frac{KS}{K_s + S} = \frac{\Delta S}{X\Delta t} \\ &R_{\text{net}} = \frac{\Delta X}{\Delta t} = \frac{KS}{K_s + S} XY - K_d X \end{aligned}\right\} \Rightarrow \frac{1}{\Delta t} = \left(\frac{\Delta S}{X\Delta t}\right) Y - K_d \tag{13}$$

Therefore, it is possible to obtain Y and K_d by plotting ($1/\Delta t$) vs. ($\Delta S/X\Delta t$).

Mass balance around the whole box could be simplified as follows:

$$\frac{dC}{dt} = r_C \tag{14}$$

Using the above information this equation can be written as follows:

$$\mathrm{r}_s r_C = \frac{dS}{dt} = \frac{KS}{K_s + S} X \tag{15}$$

$$r_X = \frac{dX}{dt} = \frac{KS}{K_s + S} YX - K_d X \tag{16}$$

The integration of the above equations would result in the following equations:

$$X = X_0 \exp\left(\int\left(\frac{KS}{K_s + S} Y - K_d\right) dt\right) \tag{17}$$

$$S(t) = S_0 - X_0 \int_0^t \left[\frac{KS}{K_s + S}\left[\exp\left(\int_0^t \left(\frac{KS}{K_s + S} Y - K_d\right) dt\right)\right]\right] \tag{18}$$

Numerical methods were used to solve these equations.

3. Results and discussion

Measurement results in the 18-day treatment period are shown in Fig. 2. It can be seen in Fig. 2 that soil around Sarkhun refinery has a high capability to remediate hydrocarbon contaminant and can reduce the amount of pollutants only by providing proper conditions such as aeration, moisture content control and sufficient nutrient addition. Also, during the 18-day treatment period in laboratory scale, removal efficiency was at least 50% in all samples. Therefore, it is possible to reduce the TPH to the standard level in a few months treatment period. According to the results, the removal efficiency at 100:5:1 and 100:9:1 nutrient ratios was almost similar. However, this result does not mean the higher the ratio the more optimal, due to the larger nutrient requirements at higher nutrient ratio. Therefore, with respect to the economical treatment, the optimum nutrient ratio in all samples is 100:5:1.

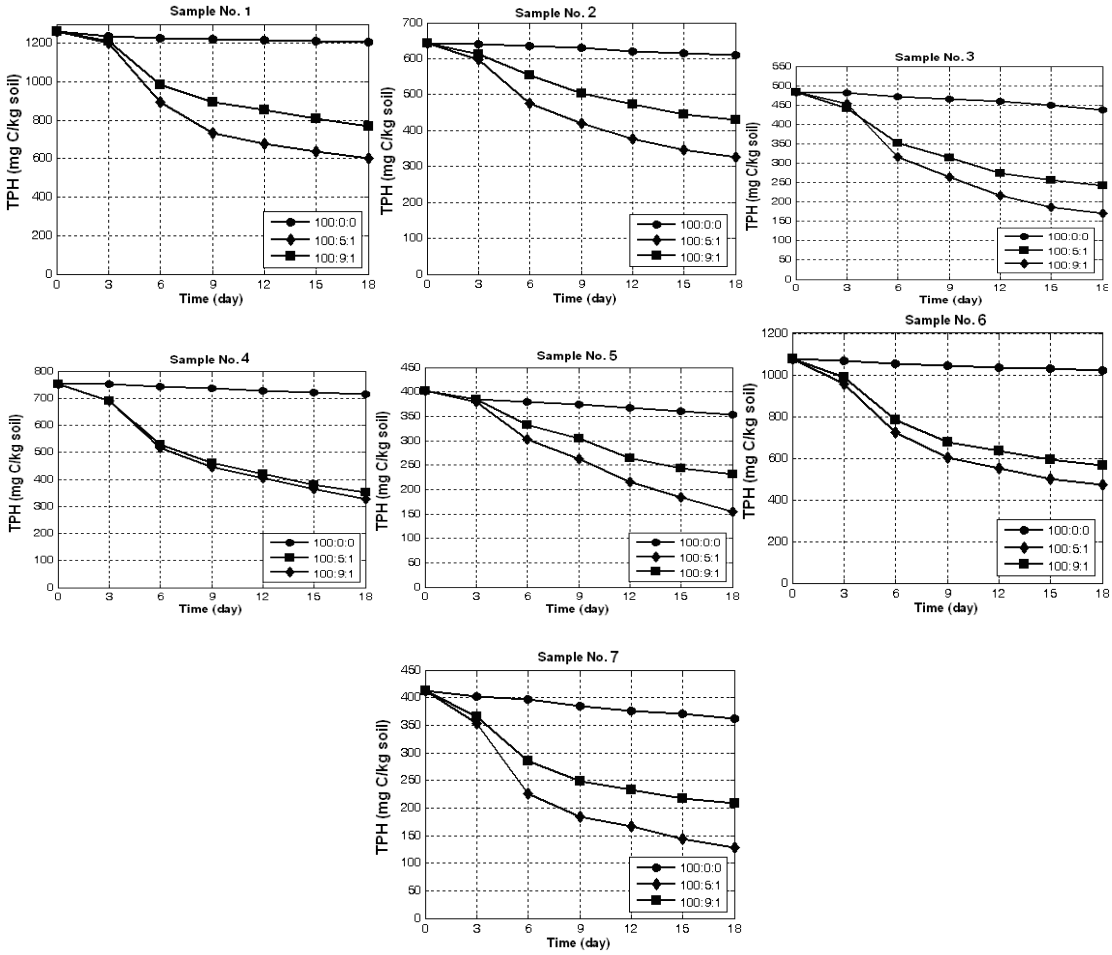

Figure 2. TPH reduction in an 18-days treatment period

According to equation 1 for a nutrient ratio of 100:5:1, B is equal to 5 and for a 100:9:1 nutrient ratio B is 9. The results obtained from this model are shown in Fig. 3.

It is obvious that the most important feature of this model is the prediction of the time needed to reach the final removal efficiency. However, this data can not be predicted from the starting times of remediation.

Extrapolating on these graphs by using the empirical model showed that at the end of the 3rd month (after a 90-day treatment period), all samples will reach the final removal efficiency, which is about 70 to 90 percent.

3-1. Modeling

In this study the limiting substrate (*S*) is TPH (mg C/kg soil). Measuring of biomass concentration (MLVSS) and TPH changes are needed for calculating the parameters of the model. In Table 1, the Biokinetic parameters in all 7 samples are represented.

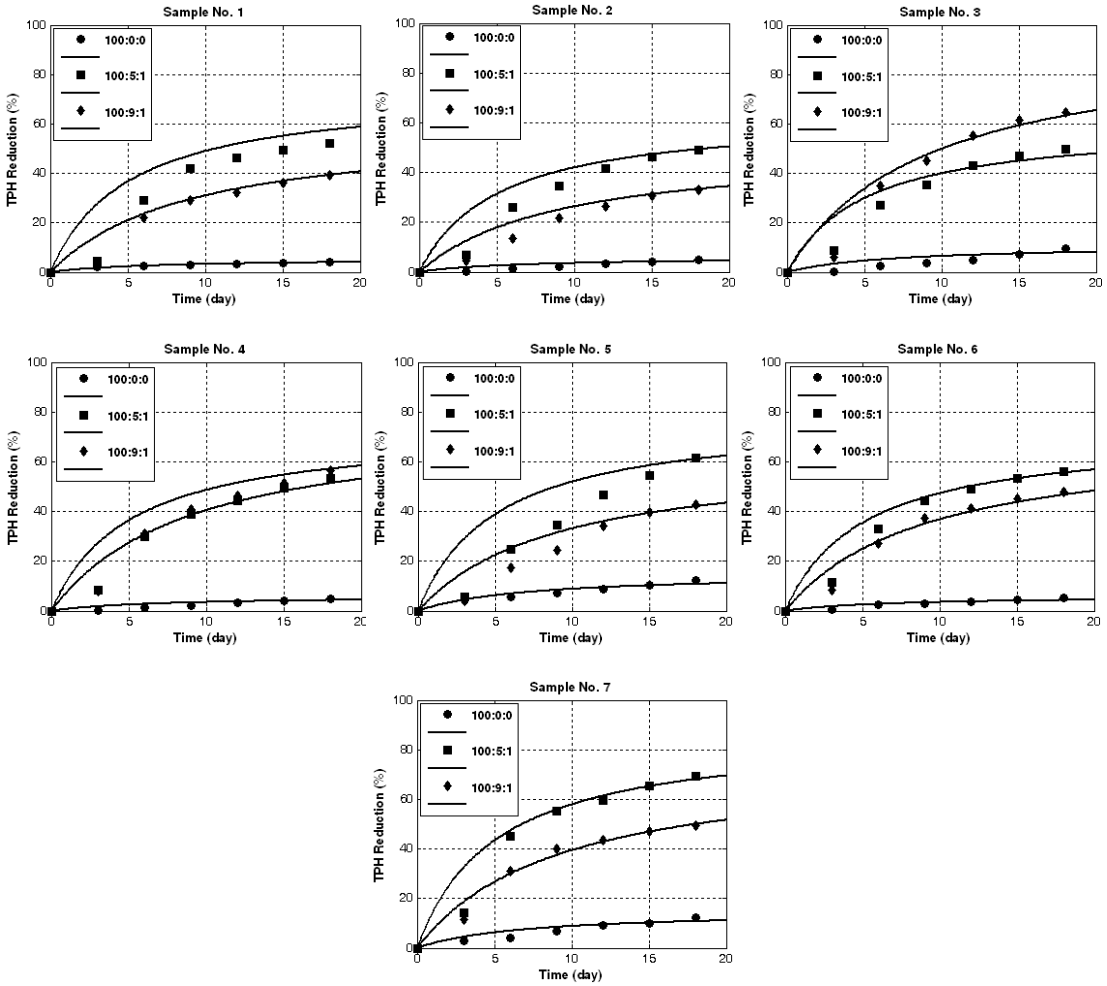

Figure 3. TPH removal efficiency, experimental data and empirical model

Table 1. Biokinetic parameters in all 7 samples

	1st sample	2nd sample	3rd sample	4th sample	5th sample	6th sample	7th sample
K (d^{-1})	3.23	3.11	3.21	3.25	3.17	3.29	3.18
K_s (mg C/kg soil)	625.43	472.91	384.22	526.97	318.48	598.16	323.35
K_d (d^{-1})	0.061	0.052	0.068	0.065	0.062	0.065	0.066
Y	0.54	0.48	0.51	0.58	0.55	0.59	0.52
μ_{max} (d^{-1})	1.74	1.49	1.64	1.88	1.74	1.94	1.65

3-1-1. Comparing experimental and modeling results

The comparison between experimental data and modeling results is represented in Fig. 4. This comparison shows that the results of modeling can simulate the behavior of bioremediation in contaminated soil with a partly suitable accuracy.

4. Conclusions

In this study, it is shown that soil around Sarkhun refinery, which has been contaminated for more than 20 years, has a high capability for bioremediation. By providing the minimum proper conditions in laboratory, mean removal percent in all seven samples was at least 50% during an 18-day treatment period. The optimum nutrient ratio was 100:5:1 for C:N:P. Also, the obtained empirical model shows that in a 90-day treatment period, it is possible to reach a final removal efficiency which is about 70 to 90%. Developing a model by taking Monod equation as biomass growth model, revealed that it is possible to predict the TPH biological removal.

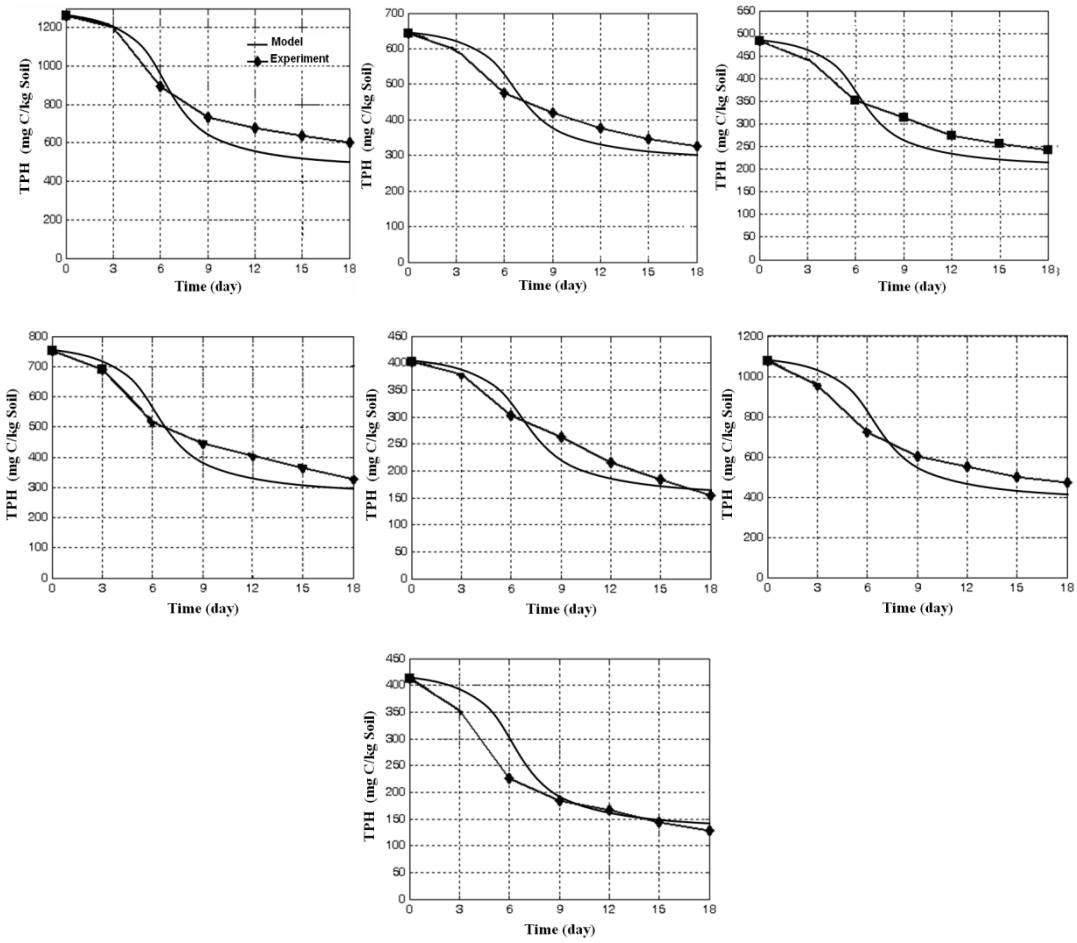

Figure 4. Comparing experimental and modeling results

Acknowledgment

The authors would like to thank the Company of Sarkhun and Gheshm gas refinery for their financial support of this study.

References

[1] Zhou, E. and Crawford, R., "Effects of oxygen, nitrogen, and temperature on gasoline biodegradation in soil", Biodegradation, 6, 127 (1995).

[2] Liebeg, E. W. and Cutright, T.J., "The investigation of enhanced bioremediation through the addition of macro and micronutrients in a PAH contaminated soil", International Biodeterioration and Biodegradation, 44, 55 (1999).

[3] Ting, H. L. Hu and Tan, H. M., "Bioremediation of petroleum hydrocarbons in soil microcosms", Resource and Environmental Biotechnology, 2, 197 (1999).

[4] Vasudevan, N. and Rajaram, P., "Bioremediation of oil sludge-contaminated soil", Environment International, 26, 409 (2001).

[5] Riser-Roberts, E., "Remediation of petroleum contaminated soil: Biological, physical, and chemical processes", CRC Press LLC, Boca Raton, FL (1998).

[6] Pirie, P., Naeimpoor, F. and Hejazi, P., "A microcosm study on P-Nitrophenol biodegradation in soil slurry by Alcaligenes faecalis: Plackett-Burman design", Iranian Journal of Chemical Engineering, 8, 2 (2011).

[7] FRTR, "Remediation technologies screeening matrix and reference guide", Ver. 4.0 (2007), http://www. frtr.gov.

[8] Pankrantz, T. M., "Environmental engineering dictionary and directory", CRC Press, Boca Raton, FL (2001).

[9] Margesin, R. and Schinner, F., "Laboratory bioremediation experiments with soil from a diesel-oil contaminated site – significant role of cold-adapted microorganisms and fertilizers", Chemical and Biotechnology, 70, 92 (1997).

[10] Jackson, W. A. and Pardue, J. H., "Potential for enhancement of biodegradation of crude oil in Louisiana salt marshes using nutrient amendments", Water, Air, and Soil Pollution, 109, 343 (1998).

[11] Huesemann, M. H. and Moore, K. O., "Compositional changes during landfarming of weathered Michigan crude oil-contaminated soil", Journal of Soil Contamination, 2 (3), 245 (1993).

[12] Li, K.Y., Zhang, Y. and Xu, T., "Bioremediation of oil-contaminated soil - a rate model", Waste Management, 15, 335 (1995).

[13] Braddock, J. F., Ruth, M. L., Walworth, J. L. and McCarthy, K. A., " Enhancement and inhibition of microbial activity in hydrocarbon-contaminated Arctic soils: Implications for nutrient- amended bioremediation", Environmental Science and Technology, 31, 2078 (1997).

[14] Bragg, J. R., Prince, R. C., Harner, E. J. and Atlas, R. M., "Effectiveness of bioremediation for the Exxon Valdez oil spill", Nature, 368, 413 (1994).

[15] Xu, Y., Lu, M., "Bioremediation of crude oil-contaminated soil: Comparison of different biostimulation and bioaug-mentation treatments", Hazardous Materials, 183, 395 (2010).

[16] http://www.astm.org/Standards/ D3921. htm.

[17] http://water.epa.gov/drink/ contaminants/index.cfm.