Malaysian Journal of Science (**23**): 211-216 (2004)

Bioremediation of maleic anhydride

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ABSTRACT This study was conducted to remediate the soil contamination issue identified in a petrochemical plant. Bioremediation was selected as the remedial options based upon a comprehensive Remedial Investigation/Feasibility Study (RI/FS) conducted earlier. This paper will present the findings of the bench scale bioremediation of Maleic Anhydride and its associated products, i.e. Phthalic Anhydride, Maleic Acid, Fumaric Acid and Benzoic Acid. The kinetic constant (K) and half-life of Maleic Anhydride were identified as –6.658 and 0.1 days, respectively. The K value and the half life of Maleic Anhydride are consistent with the available literature sources. The project was later scaled up for field implementation successfully.

ABSTRAK Kajian ini adalah untuk membersihkan tanah yang telah dicemar di sebuah kilang petrol kimia. Bioremediasi telah dipilih sebagai satu langkah untuk pembersihan setelah menjalankan kajian remediasi yang sepenuhnya. Kertas kajian ini membentangkan hasil kajian bioremediasi untuk Maleic Anhydride dan produk sampingannya seperti Phthalic Anhydride, Maleic Acid, Fumaric Acid dan Benzoic Acid. Kinatik konstant (K) dan separuh hayat untuk Maleic Anhydride yang didapati dalam kajian ini ialah –6.658 dan 0.1 hari masing-masing. Nilai K and separuh hayat ini adalah konsistan dengan nilai yang dicadangkan dalam sumber buku rujukan yang lain. Kajian ini kemudiannya telah diimplimentasikan dengan skala yang sebenarnya.

(Soil contamination, bioremediation, maleic anhydride, kinetic constant, and half life.)

INTRODUCTION

Soil contamination occurred due to the improper storage and leakage of waste by-product in a petrochemical plant. This remediation project is part of the remediation efforts that was ultimately aims to clean-up the impacted soil at the facility. The main objective of this project was to assess the feasibility and treatability of bio-remedial process for the impacted soil. As an immediate response to the contamination, the facility has subsequently conducted an initial investigation in a grid pattern at 1 foot to 4 feet below ground surface (bgs.) at the impacted area. Soil samples were collected and analyzed in the laboratory for Maleic Anhydride and Fumaric Acid.

BACKGROUND

During the project, existing site data were collected, reviewed and compiled primarily for further interpretation. Scope of work performed were to interpret and mapping of the residue concentrations in aerial and vertical extents over several sampling layers to delineate the extent of the contamination and estimate the volume of impacted soil. The largest impacted planar was at 1 feet bgs. with the area of 802 m^2 and the total volume of impacted soil was approximately 607 m^3. Based on the sampling events conducted, the primary Chemicals-of-Concern (COC) were identified based on exceedance of either the Netherlands' Dutch Intervention Values (DIV) or the US-EPA Preliminary Remediation Goals (PRGs) standards. Basically, the COC mainly consists of metals and semi-volatile organic compounds (SVOCs). Due to metals concentrations were generally detected at higher concentrations in background soil sample than samples from impacted area. It was concluded that the metals detected in the impacted soil samples are attributed to the natural occurring minerals in the geologic formation.

There were four types of SVOCs detected at high concentrations in soil at impacted area; one of the highest detected was Maleic Anhydride ($C_4H_4O_4$). The concentration detected was 223,000 mg/kg compared to PRG value of 88,000 mg/kg.

Table 1. Concentration of COCs Compared to PRG.

Matrix	Parameter	Lab. Results	Ref. Values	Int. Standards
Soil (mg/kg)	Maleic Anhydride	223,000	88,000	PRG
	Phthalic Anhydride	0.07	100,000	PRG
	Maleic Acid	20,200	NA	NA
	Fumaric Acid	14,700	NA	NA

NA - Not Available
PRG - Preliminary Remediation Goals

However, Phthalic Anhydride was detected at concentration well below the PRG value. Therefore, it is not considered as one of the COC, but since Phthalic Acid is one of the relevant degradation products of Phthalic Anhydride and it is regulated under PRG, it was added in the monitoring list of this project. Besides that, Benzoic Acid was added in this project as a monitoring parameter due to local authority's request.

Based on the physical and chemical properties of Maleic Anhydride that it's readily biodegradable, bio-treatability studies has been selected as the treatment process for investigation in this study. Tests were designed to allow evaluation of the biodegradation rates under different bench-scale treatment conditions. In favourable conditions, micro organisms are capable of degrading organic compound into carbon dioxide and water (under aerobic condition) or organic acids and methane (anaerobic). The laboratory scale experiments conducted included:

i) Control soil pile (CP) which soil matrix was neutralized with lime;
ii) Treatment pile 1 (TP1) which soil pile was neutralized with lime, and mixed with nutrients.
iii) Treatment pile 2 (TP2) which soil pile was neutralised with lime, mixed with nutrients and equipped with mechanical aeration.

The COCs monitored throughout the treatability study were Maleic Acid, Maleic Anhydride, Phthalic Anhydride, Phthalic Acid and Benzoic Acid. The physical parameters such as pH, moisture content, temperature and oxidation reduction potential (ORP) were also monitored. In addition, the microbial total plate count was also analysed to characterize the microbial activities in the soil pile.

METHODOLOGY AND APPROACHES

Representative composite soil samples were collected using a grid system by drilling equipment. Depth discrete samples were collected at 0.5 m intervals to their respective depth of 3 to 4 m bgs. A total of 300 kg of soil sample were collected. Before soil piles were constructed, the soil sample will first neutralize to pH around 7 by using lime.

The soil samples were divided equally into three portions of 100 kg each:-

a. For the first soil pile, CP, no addition of mixing materials; and
b. For the other two soil piles, TP1 and TP2, mix the neutralized contaminated soil with nutrients (chicken faeces) at recommended soil void volume (sawdust).

The carbon source and nitrogen source ratio was 1: 30 while the soil void ratio was at 30 percents based on volume basis. Two-inch diameter slotted perforated pipe, mechanical blower and electrical control panel were installed at TP2 for mechanical aeration. The three soil piles were monitored with a sampling program and samples were sent for laboratory analysis according to testing methods in Table 2.

Biodegradation kinetic rate (abbreviated as *K*) for the three experimental piles were evaluated by Michaelis Menten Equation as shown in equation (1). The derivative is a negative number because the concentration of the contaminant, S, was decreasing:

$$\frac{dS}{dt} = -\frac{k_0 SX}{K_M + S} \qquad (1)$$

Where;

Table 2. Laboratory Testing Method.

Parameters	**Test Method**
Total Plate Count	APHA 9215D
Phthalic Anhydride	Finger printing analysis SW 846 8270
Benzoic Acid	Finger printing analysis SW 846 8270
Phthalic Acid	Finger printing analysis SW 846 8270
Maleic Acid	High Performance Liquid Chromatography
Maleic Anhydride	High Performance Liquid Chromatography

X = concentration of microbes, mass/volume;
X = concentration of microbes, mass/volume;
k_m = maximum utilization coefficient, maximum rate of substrate utilization at high substrate concentration, mass substrate/mass microbes-day;
K_M = half-velocity coefficient, also referred to as the Michaelis-Menten coefficient, mass/volume;
K_0 = the maximum rate constant; and
S = concentration of the rate-limiting substrate, mass/volume.

Microbial growth is defined as microbial specific growth rate, μ derived by using Monod equation governs by equation (2):

$$\mu = \mu_m \frac{S}{K_S + S} \qquad (2)$$

Where:

μ = specific growth rate, time^{-1}
μ_m = maximum specific growth rate, time^{-1}
S = concentration of the rate-limiting substrate, mass/volume.
K_S = half-velocity constant, substrate concentration at one-half the maximum growth rate, mass/volume

The Monod model applies to soluble contaminants and heterogeneous system, where kinetic rate is a linear function of the number of active sites on contaminants and non linear with the microbial concentration. Upon development and combination of Michaelis Menten and Monod understanding, Van Uden expressed that the net specific growth rate is given by equation (3).

$$\mu^{'1} = \mu_m \frac{S}{K_S + S} - k_d \qquad (3)$$

Where;

$\mu^{'1}$ = net specific growth rate, time^{-1}
k_d = endogenous decay co-efficient, time^{-1}

Half-life usually symbolized by $t_{1/2}$, it is the time required for [S] to drop from its initial value $[S]_o$ to $[S]_o/2$. It is simply the time required for half of the amount originally present to react. At the end of one half-life, 50% of the original atoms or molecules remain.

RESULTS AND DISCUSSIONS

The physical parameters monitored compared with the recommended ranges are presented in Table 3. The biodegradation rates for the three experimental piles were evaluated by first-order kinetic. The calculated K values for Maleic Anhydride, Maleic Acid and Benzoic Acid for the three experimental piles are presented in Table 4.

Due to the presence of secondary compounds generated during the degradation process, the lag. time for the generation of these secondary compounds (Maleic Acid and Benzoic Acid) were evaluated. The lag time for these two secondary compounds to reach their maximum

Table 3. Range of Physical Parameters Recorded for Experimental Soil Piles.

Experimental Piles	**Moisture Content (%)**	**Redox Potential (mV)**	**Temp. (^{0}C)**	**pH**
CP	9.2-25.8	207-287	28-35	3.80-8.17
TP1	6.6-44	82-203	27-41	4.87-8.46
TP2	3.6-36	92-201	26-38	6.74-8.94
Recommended Range	20-40	-240 - +800	<40, 40-70, >70	6-8

Table 4. Biodegradation Rate for Experimental Soil Piles.

Experimental Piles	Biodegradation Rate, K (unit less)		
	Maleic Anhydride	Maleic Acid	Benzoic Acid
CP	-0.592	-0.010	-0.020
TP1	-5.935	-0.026	-0.029
TP2	-6.658	-0.014	-0.044

Notes: Negatives sign of K values indicative of decreasing concentrations over time.

concentrations are presented in Table 5

Experimental pile TP2 has the shortest lag time for Maleic Acid (30 days), while both the TP1 and TP2, have the shortest lag time for Benzoic Acid (10 days). The half-life calculated for Maleic Anhydride, Maleic Acid, and Benzoic Acid for the three experimental piles are presented in Table 6

TP1 and TP2 both exhibited the shortest half-life of 0.1 day for Maleic anhydride. TP1 has the shortest half-life for Maleic acid, and TP2 has the shortest half-life for Benzoic acid. In general, TP2 is the optimal experimental pile with fastest degradation process for Maleic Anhydride and Benzoic Acid; while TP1 is the optimal experimental pile for Maleic Acid, based on the degradation rate (K), lag time, and half-life obtained from the three experimental piles. The microbial specific growth rates for the three experimental piles were derived by using Monod equation, whereby the bacteria count (colony forming unit per milli-litre, CFU/ml) were tabulated against time (Table 7). The specific growth rate (μ) were derived and presented in Table 8 and Figure 1.

Table 5. Lag Time for Experimental Soil Piles

Experimental Piles	Lag Time, (day)	
	Maleic Acid	Benzoic Acid
CP	39	20
TP1	37	10
TP2	30	10

Table 6. Half Life for Experimental Soil Piles.

Experimental Piles	Half Life, $t_{1/2}$ (day)		
	Maleic Anhydride	Maleic Acid	Benzoic Acid
CP	1.1	66.4	32.2
TP1	0.1	24.3	21.9
TP2	0.1	44.5	14.5

Table 7. Bacteria Count (cfu/ml) against Time.

Day	CP	TP1	TP2
1	780	1.20E+09	6.96E+07
3	690	7.92E+08	3.80E+06
5	600	4.20E+07	3.06E+08
10	3.20E+05	8.80E+08	2.34E+08
15	1.05E+08	4.12E+08	1.02E+07
20	4.56E+07	2.37E+08	1.50E+08
25	1.44E+09	2.61E+08	2.88E+08
30	2.56E+08	5.20E+06	1.93E+08
39	3.01E+08	2.95E+06	3.66E+08

Table 8. Specific Growth Rate for Experimental Soil Piles.

Experimental Piles	Specific growth rate, μ (day^{-1})
CP	1.2073
TP1	0.6085
TP2	0.4651

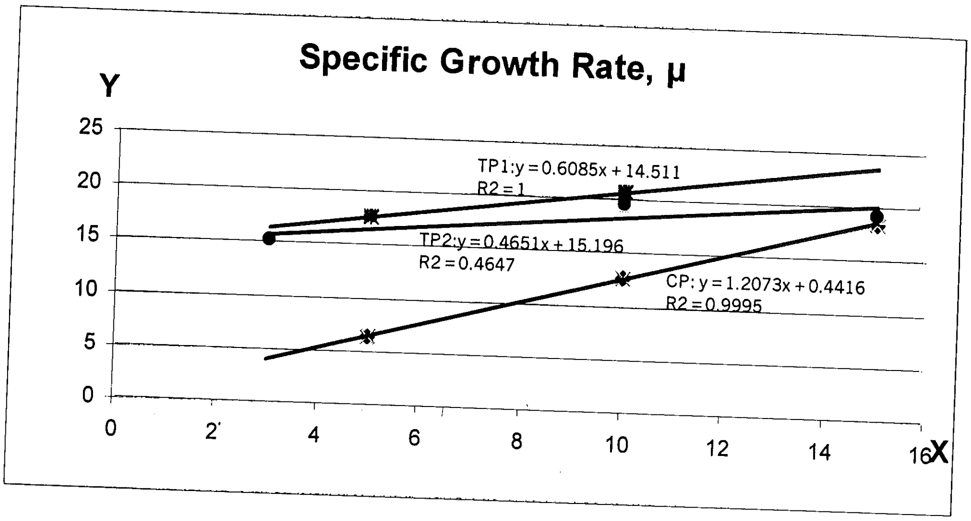

Figure 1. Specific Growth Rate for Experimental Piles Derived by Using Monod Equation.

Based on the calculation, the control pile has the highest value of specific growth rate. However, it should be noted that the microbial population was accelerated only after 20 days of experiment. Comparatively, TP1 has a slightly higher value for the specific growth rate. The range of microbial count was consistent for TP1 and TP2, which is in the magnitude of 10^6 to 10^9. Control pile has the lowest value of microbial count (in the magnitude of 10^2, Table 9). This is due to the longer lag phase experienced in the control pile. In general, TP1 supported a better environment for the growth of microbial population compared to TP2 and control.

CONCLUSIONS

Results of treatability study indicated good results were achieved in both biopiles TP1 and TP2. Generally, biopile TP2 has a higher biodegradation rate compared to biopile TP1. However, to set-up biopile TP1 was more feasible and applicable option to be implemented onsite; less resources like electricity and start-up capital cost was involved compared to setting up biopile TP2. The project was later scaled up for the field implementation successfully based on methodology and mixing ratio for treatment option TP1.

Table 9. Range of Microbial Count for Experimental Soil Piles.

Experimental Piles	Range of Microbial Count, cfu/ml*
CP	$6.00 \times 10^2 - 1.44 \times 10^9$
TP1	$2.95 \times 10^6 - 1.20 \times 10^9$
TP2	$3.80 \times 10^6 - 3.66 \times 10^8$

* Note: colony forming unit per millilitre.

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