

**UNIVERSITI TEKNOLOGI MARA**

**BIOREMEDIATION OF  
POLYCYCLIC AROMATIC  
HYDROCARBONS IN  
ARTIFICIALLY CONTAMINATED  
SOIL**

**NOR AMANI FILZAH MOHD KAMIL**

Thesis submitted in fulfillment  
of the requirements for the degree of  
**Doctor of Philosophy**

**Faculty of Civil Engineering**

**October 2014**

## CONFIRMATION BY PANEL OF EXAMINERS

I certify that a panel of examiners has met on 27th Mac 2014 to conduct the final examination of Nor Amani Filzah Binti Mohd Kamil on her Doctor of Philosophy thesis entitled "Bioremediation of Polycyclic Aromatic Hydrocarbon In Artificially Contaminated Soil" in accordance with Universiti Teknologi MARA Act 1976 (Akta 173). The Panel of Examiners recommends that the student be awarded the relevant degree. The panel of Examiners was as follows:

Mohd Fozi Ali, PhD  
Faculty of Civil Engineering  
Universiti Teknologi MARA  
(Chairman)

Ramlah Mohd Tajuddin, PhD  
Associate Professor  
Faculty of Civil Engineering  
Universiti Teknologi MARA  
(Internal Examiner)

Mohd Razman Salim, PhD  
Professor  
Faculty of Civil Engineering  
Universiti Teknologi Malaysia  
(External Examiner)

Rheo B. Lamorena-Lim, PhD  
Associate Professor  
Institute of Chemistry  
University of the Philippines  
(External Examiner)

**SITI HALIJJAH SHARIFF,**  
PhD  
Associate Professor  
Dean  
Institute of Graduates Studies  
Universiti Teknologi MARA  
Date: 13 October 2014

## ABSTRACT

Land used for industrial facilities or for waste disposal can be contaminated. Redevelopment of contaminated land often requires unacceptable risks are assessed and managed so that the site becomes suitable for its new use. One of the major criteria to be satisfied before such land can be reused is to ensure that remediation is carried out to remove all health hazards. One of the treatment methods that can be adopted involves bioremediation processes. Bioremediation process is considered to be environmentally sound and economically feasible. Other process, namely, physical and chemical processes require high energy, high costs and produce secondary contaminants that are more toxic if not completely degraded. This study is carried out to provide a better understanding on application of potential bacteria under non-indigenous condition, i.e., isolate from sludge and inoculate into contaminated soil. Many studies were conducted under indigenous and semi-indigenous condition. However, there are limited reports on studies under non-indigenous condition, particularly under aerobic condition. This study is also conducted to evaluate the effectiveness of potential bacteria and to established a mathematical model for PAHs biodegradation. The organic contaminants focus in this study is Polycyclic Aromatic Hydrocarbons (PAHs) and sand was selected as the soil media. PAHs are one of the most widespread organic contaminants and known to be highly toxic, mutagenic and carcinogenic. The objectives of this study are (i) a) establishment study parameters through characterization, (ii) biodegradation of PAHs, (iii) evaluation on effectiveness of PAHs biodegradation and (iv) model development. The methodology of this study consisted of four stages. The first stage involves determining the type of micropollutant that are present in the soil through characterization of soil taken from the Kubang Badak landfill. The second stage involves three phases of experimental works, namely, growth curve based on turbidity, biodegradation and survival of the strains in contaminated sand. The third stage involves five experimental works based on different factors, i.e., pH, temperature, phenanthrene concentration, bacteria number and heavy metals concentration. The final stage is to establish the kinetic parameter and the mathematical model for the biodegradation process. Results from stage 1 showed that phenanthrene is the most abundant PAHs in the soil samples, followed by anthracene. For heavy metals, zinc showed the highest mean concentration in the soil samples. Based on results in stage 2, a bacterial strain, namely, *Corynebacterium urolyticum* isolated from municipal sludge shows the most potential as PAHs degrader. The evaluation of most potential strain is based on four factors, namely, day of inoculation, degradation rate, bacteria survival and degradation rate per colony. Results in stage 3 showed that the optimum condition occurred at pH 7, temperature of 30°C, initial phenanthrene concentration of 500 mg/kg, bacteria number of  $10^9$  cfu/g soil and samples without the addition of heavy metals. The final stage involve development of a mathematical model based on Monod kinetic. Kinetic parameters,  $q_{max}$ ,  $X$  and  $K_s$ .  $q_{max}$  and  $K_s$  were observed to increase when the initial phenanthrene concentration increases. The Monod equation modified by Lawrence and McCarty (1970) was further enhanced in this study. As conclusion, understanding on application of potential PAHs degrading bacteria under non-indigenous condition has been evaluated in this study.

## TABLE OF CONTENT

	<b>Page</b>
<b>CONFIRMATION BY PANEL OF EXAMINERS</b>	ii
<b>AUTHOR'S DECLARATION</b>	iii
<b>ABSTRACT</b>	iv
<b>ACKNOWLEDGEMENT</b>	v
<b>TABLE OF CONTENT</b>	vi
<b>LIST OF TABLES</b>	xi
<b>LIST OF FIGURES</b>	xiv
<b>LIST OF SYMBOLS</b>	xviii
<b>LIST OF ABBREVIATIONS</b>	xix
<b>CHAPTER ONE: INTRODUCTION</b>	
1.1. Background of Study	1
1.2. Problem Statement	5
1.3. Objective of Study	6
1.4. Scope of Work	6
1.5. Significance of Study	7
1.6. Concluding Remark	9
<b>CHAPTER TWO: LITERATURE REVIEW</b>	
2.1 Introduction	10
2.2 Organic Contaminants	10
2.2.1 Polycyclic Aromatic Hydrocarbon (Pahs)	11
2.3 Soil Media	15
2.3.1 Contaminated Soil	16
2.3.2 Contributor of PAHs To Soil Contamination	16
2.3.3 Management of Closed Landfill Sites	19
2.4 Degrading Agent	20
2.4.1 Bioremediation of Soil's Degrading Agents	21
2.5 Remediation Practices For Contaminated Land	26
2.5.1 Physical Process	26

# CHAPTER ONE

## INTRODUCTION

### 1.1 BACKGROUND OF STUDY

Land is a limited and valuable resource. Land used for anthropogenic activities are likely to end up being contaminated. Contaminated land can be simply defined as the presence of foreign elements, compounds or energy on the natural soil environment that present unacceptable risks to human and the environment.

Source of contaminants may result either from natural or anthropogenic activities. For natural activities, the source may come from plant fossilization and thermal geologic reactions. On the other hand, for anthropogenic activities, spillages on soil surfaces or leakages from underground storages are commonly the source of contaminants (Aikman et al., 2007). The contaminants are capable of migrating from the point of spillage or leakage thus introducing a more widespread contamination in the soil matrix and the underlying groundwater. In addition, soil can also be contaminated through agricultural activities where excessive fertilizer and pesticides are used (Wang and Keller, 2007). Furthermore, bad engineering practices in waste management, such as, direct discharge of industrial effluent on soil surface and poor leachate management at landfills also contribute to contaminated land (Collins et al., 2002; Adeniyi et al., 2008).

Contaminated land can have adverse impact on human health and ecosystem wellbeing. The impact on human is through ingestion, inhalation or skin absorption with the contaminated land (Lim et al., 2008). Plants can also be contaminated when grown on contaminated land as they take up the contaminants through the root systems (Su and Zhu, 2007). The plant which be accumulated pollutants is not suitable for human or other receptors and is possibly unsafe to dispose directly on uncontaminated land.

Remediation of contaminated land is required to minimize the adverse impact to human and the environment. Remediation of contaminated land can be applied based on physical, chemical and biological process. Among these processes, biological