CHAPTER 1

INTRODUCTION

1.1 Statement of problem

Hexachlorobenzene (HCB) was originally introduced in the 1940's as a fungicidal seed treatment. As far as can be determined, this practice was terminated decades ago in many countries including Thailand. Using HCB as a pesticide was banned in Thailand in 1980 (Buxton, 2001). Even it was prohibited to import HCB in Thailand but the problem of this hazardous chemical still being existed. It was formed as a byproduct during the manufacture of solvents, other chlorine-containing compounds, and pesticides. Land or farms that used to be applied with pesticide or fungicide containing HCB in the past have been contaminated with HCB because of its long-range transport, deposition and persistent. HCB releases also typically include: emissions from incineration; leachate from hazardous wastes (and sometimes other) landfill sites; emissions from various industrial sources and even effluents from municipal wastewater treatment plants.

The problem of hexachlorobenzene stated in Thailand has been found by Greenpeace. It is one of 12 persistent organic pollutants (POPs) that banned by the United Nations. There is the evidence of Hexachlorobenzene in water and sediment samples taken from Klong Hua Lam Poo. It is the canal, which runs next to Bang Pho Industrial estate, a community of thousand people. It was expected that HCB would be found in sediment samples from the mouth of Bang Pakong River, which is said to be highly contaminated. There is still lack of data on this chemical in the developing world (Bangkok Post, 2001). There is not yet a reported case of people harmed by this chemical, nor a serious study on it, in Thailand.

Persistent Organic Pollutants (POPs) are chemical substances that persist in the environment, bioaccumulate through the food web, and pose a risk of causing adverse effects to human health and the environment. Twelve pollutants have been considered in POPs list are aldrin, endrin, dieldrin, chlordane, heptachlor,

hexachlorobenzene, mirex, dichlorodiphenyl trichloroethane (DDT), dioxin, polychlorinated biphenyls (PCBs), furans and toxaphene. Most of these are used in agriculture. Even long after their use has been discontinued, these chemicals remain in soils and sediments where they can enter the food chain directly or percolate down to the water table. Once in the groundwater, these pollutants can enter drinking water wells and cause health problems. With the evidence of long-range transport of these substances to regions where they have never been used or produced and the consequent threats they pose to the environment of the whole globe, the international community has now, at several occasions called for urgent global actions to reduce and eliminate releases of these chemicals.

Thai government has concerned about this problem by signing Stockholm Convention on Persistent Organic Pollutants: POPs at United Nation Head Office, New York City, USA (International Legally Binding Instrument for Implementing International Actions on Certain Persistent Organic Pollutants (POPs)) (Pollution Control Department, 2002). The cabinet resolution agree to accept Thailand to be a new member of Stockholm Convention on Persistent Organic Pollutants (May 14, 2002) : POPs via the progressing of foreign minister at United Nations headquarter, New York, United States by May 22, 2002 and accept the principal of covenant's ratification. In order to be a complete member, Thailand has to be practically prepared and also qualified from minister of pollution control department, science, technology and environment department and related departments. In addition, the committee should be set up for creating the code of practice abided by specification and commitment, the committee will set the duty for every department.

Traditional clean-up technique of chlorinated chemicals was physical process such as pump-and-treat but may be successful for removal of nonsorbing lesser chlorinated compounds, their time efficiency and cost effectiveness decreased rapidly with increasing content and molecular size due to increasing mass transfer limitations (Adriaens and Vogel, 1995). As the high chlorine content of hexachlorobenzene, it was not generally eliminated unless followed by a post-treatment process, therefore, biological degradation had significant potential to detoxify chlorinated compound.

Biodegradation was being studied as a potential transformation of chlorinated compounds to lesser-chlorinated compounds. Especially, the reductive dechlorination properties of chlorinated benzene usually takes place in reduced environments such as deep soil and sediments. Although, microbial reductive dechlorination of HCB has been well observed and documented, but, there's lack information of HCB remediation in Thailand.

1.2 Objectives

The goal of this research is to increase the rate of anaerobic biodegradation of hexachlorobenzene by examining the potential of mixed microbial species supplemented with various substrates. Homogenized granular sludge from UASB reactor was used as inoculum without acclimation in order to dechlorinate hexachlorobenzene contaminated in sediment in the combination with five types of substrates. It would be valuable data if dechlorinating ability of unacclimated sludge were observed. Because in the real situation, acclimation of sludge seed was not quite practical when large amount of sludge is needed for remediate contaminated site. The ratio between sludge and contaminated sediment quantity was also interesting with respect to cost effective. Therefore, specific objectives are:

1. To investigate the HCB dechlorinating ability of unacclimated microorganism from anaerobic reactor.
2. To study the role of carbon source and sludge:sediment ratio to the dechlorination of HCB in contaminated sediment.

1.3 Scope of study

Sample collection and preparation: The sediment was sampled from a canal run through Suanthonburirom Park, Bangkok. This sediment sample represents as contaminated sediment that will be studied. The sample was collected with a stainless steel grab sampler and added to plastic carboys with minimum headspace and analyzed to determine hexachlorobenzene background concentration. The sediment was dried, ground and sieved through 1.4-mm screen.

Condition: The experiment is carried out in 20-ml plastic syringes and kept in darkness at room temperature and pH of 6.5-7.5.

Organic substrates: Five groups of organic substrates (with COD concentration 3000 mg/l each), which are intermediates from anaerobic degradation was added to the soil samples as carbon-sources for biomass. They were glucose, lactate, ethanol, formate and the combination of acetate, butyrate, and propionate (2:1:1) (because of these three substances usually present together). Alkalinity was added to maintain pH at 6.5-7.5.

Anaerobic biomass: Granular sludge seed was derived from a UASB-wastewater treatment plant of Serm Suk Public Co., Ltd.

Nutrients: Yeast extract was fed as the culture medium in the combination with various inorganic nutrient media , see Table 3.1.

The quantification of Hexachlorobenzene: Pre-study of extraction efficiency were done six times for every single sample by adding hexane and shake for two hours. But, it was observed that the extraction results from the fourth, fifth and sixth times did not appear to be significantly higher than that of after the third time of extraction (Appendix C). Thus, only three times of extraction were done for all sample of experiment. Then, the extract was filtered and injected to Gas Chromatography for analysis.

1.4 Methodology

All experiments were done as shown in Table 1.1. The experiment for the role of c-source was conducted by varying four types and one group of c-source at consistent sludge to sediment ratio of 50:50 and controlled with no c-source addition. The role of sludge to sediment ratio was conducted by varying two ratios of 20:80 and 50:50 and both supplemented with glucose and controlled with no c-source addition. Only two sludge to sediment ratios were examined because sludge quantity should not greater than that of contaminated sediment.

Table 1.1 Experimental design

c-source / Sld:Sed*	Glucose	Ethanol	Formate	Lactate	a/b/c**	control
20:80						
50:50						

* Mass of Sludge: Sediment (gram wet weight: gram dry weight)

** acetate/butyrate /propionate

Experiment for the role of c-source.

Experiment for the role of Sludge: Sediment with glucose as c-source.

Experiment for the role of Sludge: Sediment without c-source.