ANAEROBIC BIODEGRADATION OF ALDRIN AND DIELDRIN BY MICROORGANISMS

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Introduction

Organochlorine pesticides are well known for their potential toxicity, highly hydrophobic, persistence and lipophilicity[1]. In the past decades, organochlorine pesticides have been extensively used to improve and control the agricultural products quality. Such as aldrin and dieldrin, which belong to the persistent pesticides in the environment and were classified as category B2 carcinogens by the US Environmental Protection Agency (EPA) in 1987 [2]. Although many of organochlorine pesticides have been banned in nations, due to the persistence, we can still found the residue of organochlorine pesticides in the environment [1, 3, 4]. Because of the lipophilicity, organochlorine pesticides may accumulate in animals by path of food chain. According to the reason, remediation of the contaminated place is concerned by many researchers.

Microorganisms play a major part in degradation of organochlorine pesticides [5], among them, anaerobic microorganisms have been used to degrade many organochlorine compounds in the past. Previous work has shown that the dechlorination of the hexachloronorbornene moiety of the cyclodienes aldrin, dieldrin, endrin, has been illustrated, and it has been shown that a consortium of anaerobic microorganisms create conditions for the monodechlorination[6]. This result implied that microorganism from low-oxygen or anaerobic sediment can degrade aldrin and dieldrin.

In this study, we tried to use indigenous anaerobic microorganisms from river sediment to degrade aldrin and dieldrin. In other hands, three different anaerobic conditions including methanogenic, sulfate-reducing, nitrate-reducing conditions were built. In the three defined conditions, we attempt to comprehend in which condition the aldrin and dieldrin can be degraded most efficiently.

Methods and Materials

Chemicals

Aldrin[1,2,3,4,10,10-Hexachloro-1,4,4a,5,8,8a-hexahydro-1,4-endo-exo-5,8-dimethanon-naphthalene] and Dieldrin [1,2,3,4,10,10-Hexachloro-6,7-epoxy-1,4,4a,5,6,7,8,8a-octahydro-1,4,5-endo-exo-5,8-dimetha-nonaphthalene] with 99% purity were obtained from Aldrich Chemical Co. All other compounds needed in this experiment were purchased from Sigma Co. (St. Louis). Solvent used in this experiment was HPLC-graded and purchased from Merck Co.

Sampling

Er-Jen River is one of the seriously contaminated rivers located in southern Taiwan. Anaerobic sediment was obtained from the bottom of the river. Using an Ekman grab sampler to collect the river sediment below the surface 0 ~ 10 cm in July 2001. After the sediment was collected, it was stored in jars preparing for the anaerobic seed culture.

Culture medium

A 1 L serum bottle with 400 ml anaerobic medium and 300 g anaerobic sediment are used for incubating anaerobic seed culture. The anaerobic medium was consisted of (all concentrations in g/l) :

NH_4Cl, 2.7; $MgCl_{2?}6H_2O$, 0.1; $CaCl_{2?}2H_2O$, 0.1; $FeCl_{2?}4H_2O$, 0.02; K_2HP0_4, 0.27; KH_2P0_4, 0.35; yeast extract, 1.0; resazurin, 0.001. Medium pH was adjusted to 7.0 following autoclaving; 0.9 mM titanium citrate was added as a reducing reagent. The medium was modified from Chang[7]. Three defined conditions including: methanogenic conditions (20mM $NaHCO_3$ added), sulfate-reducing conditions (20 mM $NaSO_4$ added), nitrate-reducing conditions (20mM $NaNO_3$ added) and inoculated control (absence of sodium sulfate, sodium nitrate or sodium hydrogen carbonate). All operations were progressed in an anaerobic glove box filled with N_2 (85 %), H_2 (10 %), and CO_2 (5 %) gases.

Batch experiments

Aldrin and Dieldrin degradation test were performed using 125 ml serum bottles containing 45 ml medium, 5 ml anaerobic seed culture, and spiked 2 ppm aldrin and dieldrin. Serum bottles were sealed with butyl rubber stoppers and capped with aluminum foil. All the bottles were incubated without shaking at 30? in darkness in order to prevent photolysis. All experiments were performed in triplicate.

Analysis

Extraction was performed in 15 mL tubes, by adding 2 mL *n*-hexane and shaking for 1 minute, after the initial *n*-hexane layer was removed, culture was extracted with two additional *n*-hexane treatments. The concentration of aldrin and dieldrin were measured with a gas chromatograph (Hewlett-Packard 6890 series N) equipped with an electron capture detector (ECD) and a HP 1 fused silica capillary column (film thickness, 0.33 mm; inner diameter, 0.25 mm; length, 30 m). Nitrogen was used as both a carrier and make-up gas with flow rates of 2.5 mL/min (20:1 split ratio). The column temperature program was set at 170° in initial for 2 min, then increased by 2.5 ml/min to 210°, where it was maintained for 2 min before being increased by 10 ml/min to 250°, where it was maintained for 5 min. Injector and detector temperatures were set at 250 and 300°, respectively.

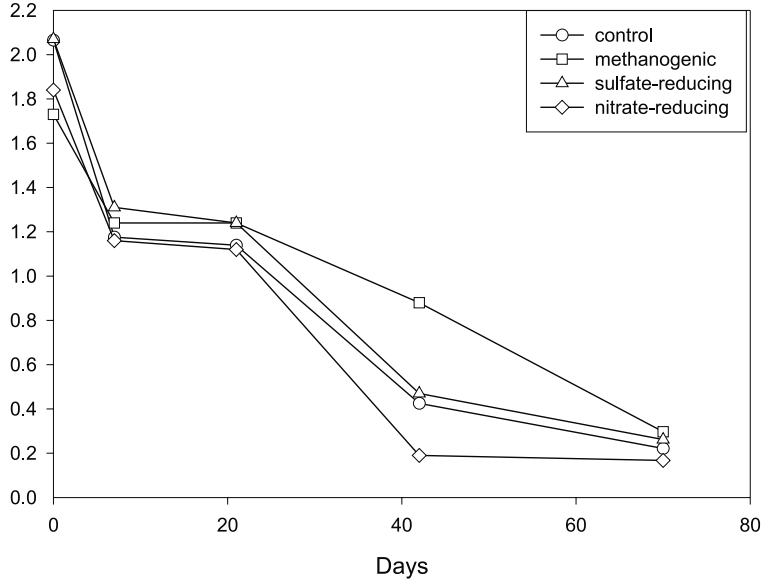

Fig 1. Degradation of Aldrin by anaerobic microorganisms

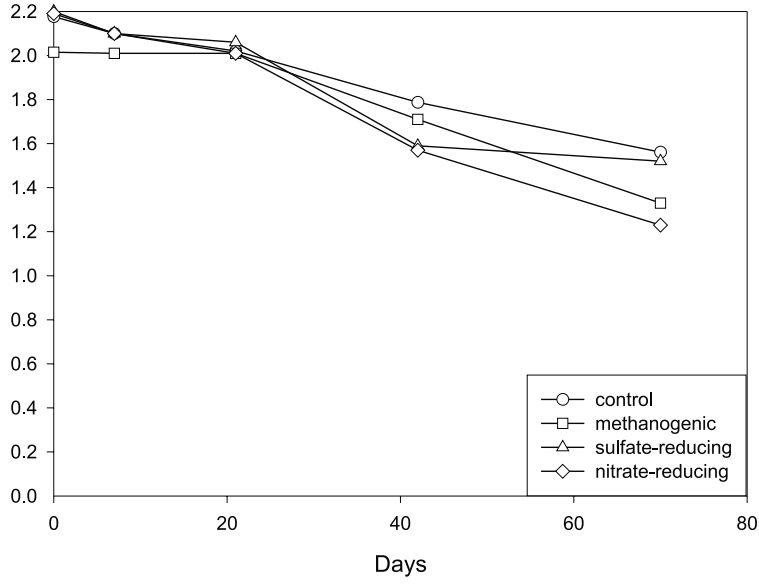

Fig 2. Degradation of Heptachlor by anaerobic microorganisms

Result and discussion

Aldrin and dieldrin belong to persistent organochlorine pesticides, in this research, we attempt to comprehend the microbial ability to degrade aldrin and dieldrin. From the result shows in Fig 1. There are not significant different degradation rate between the three defined conditions (methanogenic, sulfate-reducing and nitrate-reducing stages) and inoculated control. Aldrin can be degraded over 70 percent during 70 days. In the other hand, the degradation of dieldrin shows a slowly transform process (Fig 2), but we can still find a different percentage of degradation in order: nitrate-reducing condition (43.8 %) > methanogenic condition (34 %)> sulfate-reducing condition (26.2 %)> inoculated control (22.7 %). In this study, we found anaerobic microorganisms can also degrade persistent organochlorine pollutants such as aldrin and dieldrin. Although microorganisms displayed a poor ability to reduce dieldrin, it might represent there is lack of microorganisms which have the ability of degraded dieldrin in the river sediment environment. From the result, the anaerobic microorganisms from river sediment in Taiwan provided the potential to degrade aldrin and dieldrin.

Acknowledgement

The reaserch was supported in part with funds provided by the National Science Council of the Republic of China (NSC -90-2317-B-002-012).

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