MUTANT STRAINS FOR BIODEGRADATION OF PCBs IN BIOREACTOR SYSTEM

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INTRODUCTION

The desirable physical and chemical properties of polychlorinated biphenyls (PCBs) allowed their widespread use in industry. During the past 45 years, the total world production of PCBs has exceeded one million tons, approximately 40% of which was released into waterways. This investigation was conducted to develop microbial systems able to resist high levels of PCB (1248 and 1254) and to use these compounds as a sole source of carbon and energy. We also examined the rate of Carbon-14-PCB (C-14-PCB) uptake and its fate in bacterial cells as well as its removal from waste using activated charcoal (AC).

METHODS

The C-14-PCB in culture and in the cell following uptake was counted using a Beckman LS-7000 liquid scintillation system. PCBs were recovered from the medium used for uptake experiments by three consecutive hexane Excess water was removed from extractions. hexane extracts by adsorption using anhydrous sodium sulfate. PCBs were assayed using a Tracor Model 560 GC equipped with a Ni-63 electron-capture detector and a Pyrex glass column (0.4 x 180 cm) containing 3% OV-1 on Chromosorb WHp (80-100 mesh) as well as J & W Fused Silica Capillary column (0.25 mm K.D. x 30 m) containing Durabond DB-5 [poly-methyl (5% phenyl) Siloxane].

Four different groups of organisms (three bacteria and one mold) were isolated from PCBcontaminated soil and sediments (in GA and PA) and identified as <u>Serratia</u>, <u>Pseudomonas</u>, Bacillus, and <u>Aspergillus</u> sp.

RESULTS

Mutant strains of these organisms were developed from 300-2000 g/ml. Figure 1 shows the effect of various concentrations on the survival of <u>Serratia liquifacions</u> indicating its resistance to high levels of PCB. Uptake experiments using C-14-PCB revealed that most of the PCB was in the lipid fraction of bactertial cell walls and membranes (Table 1). Both alcohol and alcohol-ether fractions contained most of the radioactivities indicating the bioaccumulation in the soluble proteins and lipid fractions of cells. Bioreactor (33 gal) experiments using contaminated waste (PCB 1248 and 1254) showed that <u>Pseudomonas</u> and <u>Serratia</u> sp. were able to degrade most PCB in 90-130 days with a half-life of 15 days.

PCBs were adsorbed by AC from industrial waste and recovered from the AC by repeated hexane extraction, suggesting AC adsorption as a possible method for removal of low levels of PCB from contaminated industrial waste.

CONCLUSIONS

Biochemical and pathway studies revealed that dechlorination of PCB occurred in cell wall followed by metabolism in cytoplasm via oxidative pathways to lower chlorinated and dechlorinated molecules, CO_2 and H_2O and that these organisms were able to destroy PCB at a biological halflife of 15 days.

RECOMMENDATIONS

The use of PCB-mutant strains can be of great value for the biodegradation of PCB 1254 in the contaminated wastes.

LITERATURE CITED

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- Hamdy, M. K., and Gooch, J., 1986. Update, Retention, Biodegradation and Depuration in PCB by Organisms. <u>In</u>: CRC PCBs and the Environment, Vol. II, John S. Waid (Editor). CRC Press, Inc., Boca Raton, FL. pp. 63-88.

Fractions	Serratia ^a (%)	Pseudomonas ^a (%)	Bacillus ^b (%)
Cold TCA Soluble	0.7	1.1	3.6
Alcohol Soluble	42.0	50.0	67.4
Alcohol-Ether Soluble	57.0	48.5	19.6
Hot TCA Soluble	0.1	0.3	5.3
Residual Precipitate	0.2	0.1	5.1

Table 1. C-14-PCB Distribution in Cellular Fractions of Bacteria* after 72 hr Incubation (37°C).

* Based on % total activities recovered from all fractions Average of four experiments Average of two experiments а

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0 -1 -2 LOG N/No -3 2000ug/ml 1000ug/ml 1000ug/ml -4 500 yg/ml 300µg/ml -5 -6 0 20 40 60 80 100 120 EXPOSURE TIME (min.)

The comparative effect of various concentrations Figure 1. of PCB 1254 on survival of <u>Serratia</u> <u>liquifaciens</u> in phosphate buffer incubated at $37^{\circ}C$ for 2 hr. N denotes the number of cells after exposure and N represents cell number at zero time.