REDUCTIVE DECHLORINATION OF 2,6-DICHLOROPHENOL IN AN ANAEROBIC BIOREACTOR

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ABSTRACT :
Methanogenic degradation of chlorophenolic compounds has been studied under conditions of continuous substrate feeding in an upflow sludge blanket bioreactor (operative volume, 140 cm3) using 2,6-dichlorophenol (2,6-DCP) as the model substrate.

INTRODUCTION :
Previous batch-culture experiments in this laboratory have shown the ability of methanogenic surface-layer sediment from the highly polluted Saale river to completely degrade various mono- and dichlorophenols after a certain initial acclimation period with CH4 being produced in amounts corresponding to 65-95% of the theoretically possible value (KAMINSKI et al. in preparation). In the present work, an upflow sludge blanket bioreactor has been developed to study methanogenic degradation of chlorophenolic compounds by pre-adapted anaerobic consortia from Saale river sediment under conditions of continuous substrate feeding using 2,6-DCP as the model substrate.

EXPERIMENTAL PART :
Stable 2,6-DCP-metabolizing methanogenic enrichment cultures were obtained by inoculating pre-reduced phosphate-buffered RAMM-medium (SHELTON and TIEDJE 1984) with black anoxic mud (10% w/v) collected from Saale river near Jena and subsequent stationary incubation (including periodical refeeding with 2,6-DCP as the sole organic carbon substrate) under strictly anaerobic conditions for appr. 2 years in analogy to the procedure described by KAMINSKI et al. (1990). The particulate fraction from cultures of this type was collected (under nitrogen stream) and used as the inoculum material for subsequent long-term degradation experiments in upflow reactor systems (working volume, 140 cm3) of a design shown in Fig.1. Three of such reactors were operated simultaneously with the following modifications : (i) with 120 cm3 of inoculum (bioreactor 1), (ii) with 40 cm3 of inoculum (bioreactor 2), (iii) with no inoculum material added (abiotic control; reactor 3).

2,6-DCP and its dechlorination products 2-chlorophenol (2-MCP) and phenol were analyzed by gas-liquid chromatography. For this purpose, 1-ml samples from the influent and effluent, resp., were acetylated by adding 0.5 ml 10% NaHCO3 (containing 1 mM m-cresol as the internal standard) and 50 ul of freshly distilled acetic anhydride. Then, the mixtures were extracted each into 200 ul n-hexane and the hexane phases were analyzed with a GC14A-type gas chromatograph (Shimadzu, Japan) equipped with a HP-1 (Hewlett-Packard, USA) capillary column and a flame-ionization detector. Under the conditions used, the relative retention times (compared to m-cresol) were : 2,6-DCP, 1.21; 2-MCP, 1.05; phenol, 0.88 .
Redox potential measurements were performed with a MV87-type digital pH-meter (Präzitronic, GDR) using a platinum electrode and a KE10-type (Hg/Hg2C12/KCl saturated) reference electrode.

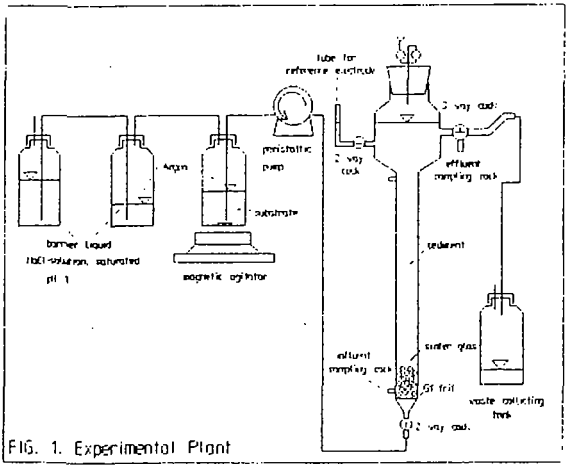

FIG. 1. Experimental Plant

RESULTS AND DISCUSSION :

Following inoculation with the particulate fraction of 2,6-DCP-metabolizing methanogenic batch-cultures, ortho-dechlorinating activity towards 2,6-DCP as well as a redox potential of less than -250 mV were maintained in both bioreactors 1 and 2 for more than 120 days under the operative conditions used. In contrast, 2,6-DCP removal in reactor 3 (abiotic control) did not exceed 5% (without formation of any dechlorination product). As shown in Fig. 2, in bioreactor 1 substrate loading rates of up to 8 mg 2,6-DCP per day at a hydraulic retention time (HRT) of 1 day were achieved at a substrate conversion efficiency of >90% . A considerably lower 2,6-DCP-degrading volumetric activity was seen in bioreactor 2 indicating that this parameter essentially depended on the inoculum size. In general, however, the specific substrate conversion rates in bioreactors 1 and 2 were nearly those calculated for the foregoing batch-culture experiments.

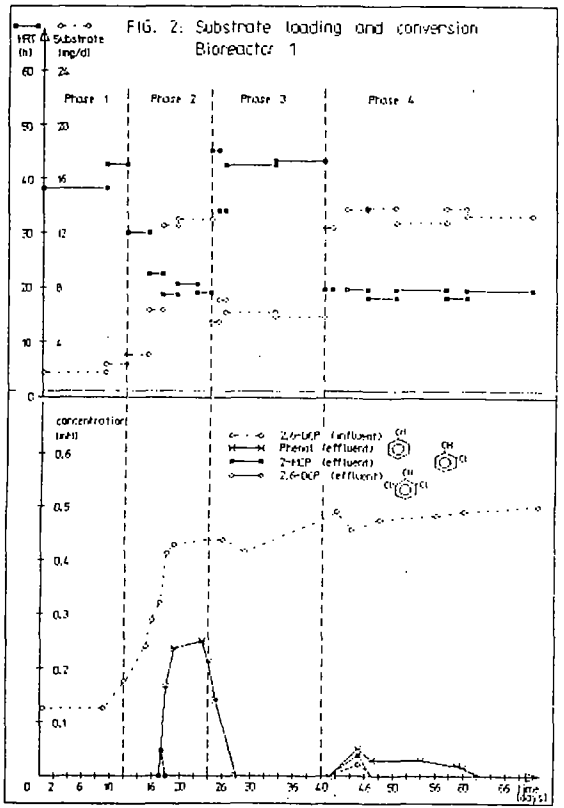
FIG. 2: Substrate loading and conversion
Bioreactor 1
HRT (h)
Substrate (mg/d)
Phase 1
Phase 2
Phase 3
Phase 4
concentration (mM)
2,4-DCP (influent)
Phenol (effluent)
2-HCP (effluent)
2,6-DCP (effluent)
time (days)

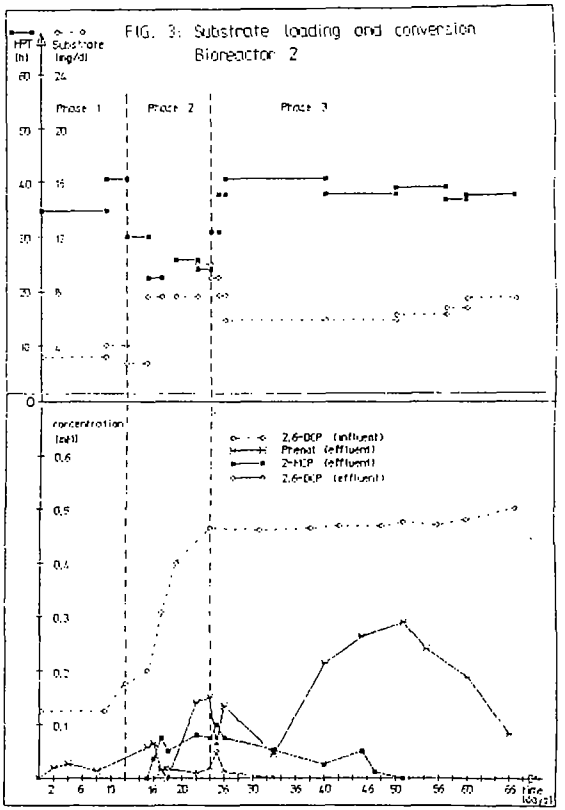
FIG. 3: Substrate loading and conversion
Bioreactor 2
HRT (h)
Substrate (mg/d)
Phase 1
Phase 2
Phase 3
concentration (mM)
2,6-DCP (influent)
Phenol (effluent)
2-HCP (effluent)
2,6-DCP (effluent)
time (days)

In both bioreactors, 2-MCP as well as phenol were found as the products of reductive ortho-dechlorination in concentrations which depended on the actual substrate loading rate.

Anaerobic biofilters represent a low-cost alternative to traditional aerobic activated-sludge systems in the context of bioremoval of riskful man-made organic compounds. The results of our model experiments clearly evidence that pre-adapted methanogenic consortia like those from Saale river sediment can be used as a special biological inoculum material to develop/initiate anaerobic treatment systems for an efficient elimination/detoxification of 2,6-DCP and other chlorophenolic compounds from industrial waste waters or contaminated groundwaters. It is intended to use the developed anaerobic upflow bioreactors for further experiments on biotransformation (i.e. reductive dechlorination) of highly chlorinated phenols and other persistent chloroorganic compounds under conditions of continuous substrate feeding by mixtures of differentially acclimated methanogenic consortia in the absence /presence of easily degradable organic carbon compounds.

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