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TOXIC EFFECTS OF PHENOLIC POLLUTANTS ON ANAEROBIC BENZOATE-DEGRADING GRANULES

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SUMMARY

Phenolic pollutants were toxic to the bioactivity of anaerobic benzoate-degrading granules. Cresols were more toxic than phenol, which in turn was more toxic than dihydroxybenzenes. The toxicity was dependent on the hydrophobicity of the functional group, but not their positions. The granules exhibited mild resistance to the toxicity of phenolic pollutants, probably because of their layered microstructure.

INTRODUCTION

The upflow anaerobic sludge blanket (UASB) process (Lettinga *et al.* 1980; Fang and Chui 1993) has been successfully commercialized in the past decade for the treatment of wastewaters from food/beverage and agricultural industries. It could also be effective for the treatment of industrial wastewaters containing aromatic chemicals, such as benzoate. Li *et al.* (to be published) demonstrated that over 97% of benzoate in wastewater could be removed by UASB process at 37°C with 9.8 hours of retention time for wastewater containing up to 6300 mg/L of benzoate, equivalent to 12500 mg/L of chemical oxygen demand (COD). On the other hand, phenol and its derivatives are common pollutants in wastewater of chemical industries. They are used for the production of a variety of specialty chemicals (Kirk Othmer, 1978). This study was conducted to examine their toxic effects on the bioactivity of benzoate-degrading UASB granules.

MATERIALS AND METHODS

Sludge granules were sampled from the reactor for bioactivity tests from an 8.5-liter

UASB reactor which had been operated using benzoate as the sole substrate (Li *et al.* to be published) at a loading rate of 10 g-COD/L•day for three months, during which the reactor consistently removed 98.5-99.5% of COD. The bioactivity of the sampled granules were measured by the rate of converting benzoate into methane in the 157 mL serum vials (Owen, *et al.*, 1979; Dolfing and Mulder, 1985). Each vial was filled with 100 mL of synthetic wastewater which not only contained 1260 mg/L of benzoate (equivalent to 2500 mg/L of COD) as substrate, plus nutrients, vitamins, trace elements, *etc.*, but may also contain individual phenolic pollutants at controlled dosage. After granules were added, each vial was capped with butyl rubber and submerged in 37°C water in a shaking bath. About 100 mg of biomass were added to each vial; the exact amount was measured by the content of volatile suspended solids (VSS), following the standard methods, at the end of each test. Transfers of wastewater and granules to the serum vials were conducted under anaerobic environment.

Biogas production was monitored every few hours using syringes; the methane contents in the biogas were measured using a gas chromatograph equipped with a thermal conductivity detector. The initial slope of specific methane production versus time represented the specific methanogenic activity (SMA) of the granules using benzoate as substrate. The SMA in each serum vial was dependent on the dosage of individual phenols. All SMA measurements were conducted in duplicates. Up to 30 vials were monitored in each batch; at least two of the 30 vials without phenols served as control. The toxicities of individual phenols at various concentrations were indicated by the decrease of SMA as compared to the controls.

RESULTS AND DISCUSSION

The specific methane production rate decreased with the increase of toxicant/biomass ratio. Figure 1 illustrates the case of phenol. The SMA of the granules was about 95% of the control when each gram of biomass was in contact with 0.45 gram of phenol. The bioactivity was reduced to 69% of the control when the granules were in contact with 1.43 grams of phenol, and decreased to nil when in contact with 2.2 grams of phenol. The IR_{50}, which represents the ratio of phenol to biomass at which the methanogenic activity of benzoate-degrading granules was 50% of the control, was 1840 mg/g for phenol, as illustrated in Figure 1. The corresponding concentration, IC_{50}, of phenol in wastewater was 1850 mg/L.

Similar plots for three dihydroxybenzenes show that the granules were somewhat more sensitive to catechol than to resorcinol and hydroquinone, the respective IR_{50} being 1890, 2340 and 2270 mg/g-VSS and the corresponding IC_{50} 2250, 2400 and 2500 mg/L. For cresol isomers, results show that the relative bioactivities of three cresols were very similar. The respective IR_{50} were 870, 950, and 925 mg/g-VSS for the ortho-, meta-, and para- isomers; the corresponding IC_{50} were 850, 925 and 975 mg/L.

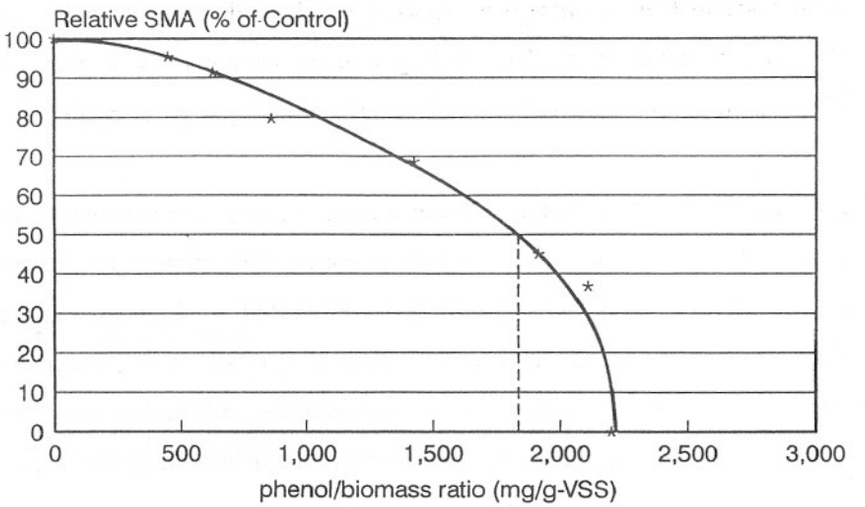

Figure 1: Inhibition of bioactivity of granules by phenol

Table 1 summarizes the IR_{50} and IC_{50} data observed for benzoate-degrading granules. Sierra-Alvarez and Lettinga (1991) investigated the toxic effect of organic pollutants on UASB granules treating distillery wastewater by measuring the reduction of the granules' bioactivity on converting acetate to methane. Their corresponding IC_{50} data are also included in Table 1 for comparison.

Table 1 IC_{50} and IR_{50} of Phenol and Phenolic Derivatives

	Benzoate-Degrading UASB Granules		Distillery UASB Granules*
pollutants	IR_{50} (mg/g-VSS)	IC_{50} (mg/L)	IC_{50} (mg/L)
phenol	1840	1850	1100
catechol	1890	2250	1813
resorcinol	2340	2400	N/A
hydroquinone	2270	2500	N/A
ortho-cresol	870	850	N/A
meta-cresol	950	925	N/A
para-cresol	925	975	568

* Sierra-Alvarez and Lettinga (1991)

Results in Table 1 shows that the chemical nature of the functional group was more critical to the toxicity than its position on the aromatic ring. Phenols with a hydrophobic

methyl functional group (i.e. cresols) were more toxic than phenol, which in turn was more toxic than the dihydroxybenzenes. This finding was in agreement with Sierra-Alvarez and Lettinga (1991), namely, the more hydrophobic the functional group the higher the toxicity.

Although phenol and its derivatives were inhibitory, this study demonstrated that their inhibitory effect was not severe for the UASB granules. The relative resistance to toxicity exhibited by the UASB granules can probably be attributed to their microstructure. Many UASB granules have a layered microstructure (MacLeod, *et al.*, 1990; Fang, *et al.*, 1994), their outer layer being mostly composed of fermentative and acidogenic bacteria which converted benzoate and other complex pollutants into fatty acids. These bacteria shielded the toxicity-sensitive methanogens in the interior of granule.

Table 1 also shows that the benzoate-degrading granules exhibited higher resistance to the toxicity towards phenols than those granules treating distillery wastewater. This was probably due to the fact that the former granules had been acclimated to an aromatic substrate, benzoate.

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