

## Effects of Suspended Solids on the Biotransformation of Acenaphthene

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Biotransformation is a major fate process for many chemicals in aquatic environments. Regulatory requirements such as the Toxic Substances Control Act (TSCA) and manufacturers' interests in the fate of chemicals they produce have created an interest in laboratory-derived biotransformation rates and their extrapolation to natural environments.

One factor currently of interest is how suspended solids may affect the biotransformation rate of chemicals. Several investigators have attempted to assess the role of suspended solids in biotransformation rate studies with varying results (Pignatello et al. 1983; Herbes 1981; Lee and Ryan 1979; Simsiman and Chesters 1975). The results of these studies indicate that no generalizations can be made about the importance of suspended solids in altering the biotransformation rate of chemicals.

Several explanations exist for the effects of sediments on biotransformation rates. Sorption onto sediments is presumed to be the controlling factor for many chemicals (Pritchard 1984, unpublished). If the chemical is reversably sorbed to solids, rate of desorption may be the rate-controlling step if water column bacteria are transforming the chemical in question. solids-associated bacteria are transforming the chemical then sorption may increase the biotransformation rate. However, there are rare examples of chemicals, such as diquat, which are not biotransformed when sorbed to solids (Simsiman and Chesters 1976). Another possibility for sediments to affect biotransformation rates would be by the addition of microorganisms associated with solid materials. If the second order model of Paris et al. (1981) the chemical under study, increases in microorganisms biotransformation rate would be expected to increase the coefficient.

Most research in this area deals with natural water-solids systems. This approach is necessary when extrapolating results to the environment. However, natural environments are complex and contain many undefined variables. This "whole system" approach does not easily lend itself to isolation and characterization of any one component or process within the system. For this reason,

and to assess the importance of suspended solids in altering biotransformation rate, a unique approach was taken to study the effects of suspended solids on water column biotransformation.

The objective of this study was to determine the effects of different types and varying levels of suspended solids on the biotransformation rate of acenaphthene. Acenaphthene was selected as the test chemical because it is a priority pollutant, was expected to be susceptible to biotransformation processes, and is a representative of polynuclear aromatic hydrocarbons, pollutants commonly found in aquatic environments. In order to isolate the effects of suspended solids on biotransformation, suspended solids sources and concentrations were varied in EPA reconstituted hard water. A "synthetic" water source was used to reduce variability in the system and to isolate the effects of suspended solids on the biotransformation rate of acenaphthene.

## MATERIALS AND METHODS

Acenaphthene (CAS #83-23-9) was obtained from the Eastman Kodak Company (Rochester, NY). Suspended solids sources were sediments from Pat Mayse reservoir (PM), a mesotrophic reservoir in Paris, Texas and Roselawn Pond (RP), an eutrophic pond in Denton, Texas. Sediments were collected using an Eckman dredge. All sediments were wet sieved through a 180 um standard mesh sieve and stored in glass containers at  $4^{\circ}$ C. Selected characteristics of these sediments (Table 1) were determined according to Black et al. (1965) and Standard methods (APHA 1980).

Table 1. Selected characteristics of suspended solids.

	Suspended Solids Source		
Characteristic	Roselawn Pond (RS)	Pat Mayse Lake (PM)	
Particle Size µm	<180	<180	
pH at 25 <sup>0</sup> C	8.04	7.36	
Percent Sand	0	12	
Percent Silt	33	22	
Percent Clay	67	66	
Percent Organic Carbon	$1.64 \pm 0.001^{1}$	$0.70 \pm 0.016^{1}$	
Percent Dry Weight	31	50	

<sup>1</sup> Standard Deviation of Replicate Measurements

Acenaphthene was dissolved in reconstituted hard water (EPA 1975) stirring in the dark for 24  $\ensuremath{\text{h}^{-1}}$  then filtered through Schleicher and Schuell #30 (0.45 micron) glass fiber filters to remove undissolved acenaphthène. The resulting solution contained mgL<sup>-1</sup> acenaphthene as determined approximately 2.0 spectrofluorometric analysis. Experimental systems consisted of reconstituted hard water with acenaphthene and nominal suspended solids concentrations of zero, 100, 500, and 750  $\,\mathrm{mgL}^{-1}$  in either 125 or 250 ml Erlenmeyer screw-top flasks with teflon liners. Test solution volumes in the 125 and 250 ml flasks were 100 and respectively. Controls consisted of autoclaved experimental systems. Incubation was at room temperature (20 to 24°C) on an Eberbach shaker table reciprocating at 140 rpm. Experiments were conducted under aerobic conditions in the dark.

At selected time intervals following initiation of an experiment three ml of solution were removed from each flask for determination of remaining acenaphthene concentrations. This was accomplished by a 1:1 extraction with pesticide grade hexane. Extraction was completed by mixing for two minutes on a thermoclyne maxi-mix. Percent recovery of acenaphthene from solutions containing suspended solids was determined to be >90%. Subsequent analysis was performed on the hexane fraction using an Aminco-Bowman spectrofluorometer (excitation  $\lambda$ =291 nm, emission  $\lambda$ =335 nm). Acenaphthene concentration was determined by comparing fluorescence to standards. Sterility of controls was confirmed at the termination of an experiment by standard pour plate techniques using 0.1% plate count agar (Difco) and 1% agar (Difco). Plates were incubated in the dark at 20°C for 120 h $^{-1}$  then examined.

Psuedo-first order biotransformation rate coefficients were calculated by plotting  $\ln(\text{Ct/Co})$  versus time (Williams et al. 1978), where Ct equals the acenaphthene concentration at time t and Co equals the acenaphthene concentration at time zero. Using this method the slope equals the negative rate coefficient -(K). Biotransformation rate coefficients were corrected for abiotic processes. Tests for statistically significant differences (alpha = 0.05) between slopes were conducted using analysis of covariance procedures (SAS 1982).

## RESULTS AND DISCUSSION

Table 2 shows the proportion of acenaphthene remaining (Ct/Co) at selected time intervals in all exposures. No clear difference in control and experimental systems containing zero suspended solids is evident after 7 d (ratio of Ct/Co in control and experimental systems at day 7=1.17). Statistically significant differences were not found between rate of loss of acenaphthene in these exposures, indicating that microbes associated with the diluent were not contributing significantly to loss of acenaphthene. Loss of acenaphthene in control and experimental systems without suspended solids is illustrated in Figure 1. A 3 to 16 fold difference exists between mean Ct/Co after 11 d $^{-1}$  in control and experimental systems with equal levels of RP sediments. However,

Table 2. Loss of Acenaphthene in biotransformation experiments.

			Ct/Co				
	Measure	nd1 C	Controls		Experimental		
Day	Solids (mg/l)	;	Standard Deviation	Mean	Standard Deviation		
2	0	0.98	0.04	0.99	0.02		
5	0	**	**	0.68	0.10		
7	0	0.74	0.23	0.63	0.09		
3	52 RP	0.85	0.13	0.59	0.07		
3	403 RP	0.69	0.36	0.24	0.03		
3	601 RP	0.97	0.02	0.24	0.02		
5	52 RP	**	**	0.42	0.17		
5	403 RP	**	**	0.20	0.05		
5	601 RP	**	**	0.13	0.02		
9	52 RP	**	**	0.10	0.04		
9	403 RP	**	**	0.04	0.02		
9	601 RP	**	**	0.04	0.07		
11	52 RP	0.65	0.12	0.14	0.12		
11	403 RP	0.46	0.34	0.14	0.08		
11	601 RP	0.67	0.22	0.04	0.01		
2	83 PM	0.94	0.09	0.84	0.11		
2	397 PM	0.98	0.03	0.70	0.22		
2	5,91 PM	0.98	0.33	0.66	0.25		
5	83 PM	**	**	0.44	0.12		
5	397 PM	**	**	0.06	0.06		
5	591 PM	**	**	0.09	0.17		
7	83 PM	0.83	0.14	0.33	0.12		
7	397 PM	0.92	0.11	0.03	0.01		
7	591 PM	0.94	0.10	0.01	0.01		

<sup>&</sup>lt;sup>1</sup>RP = Roselawn Pond Sediments, PM = Pat Mayse Lake Sediments.

statistically significant differences were found only between control and experimental systems with 601 mgL<sup>-1</sup> RP sediments. Statistically significant differences were not found between rate of loss of acenaphthene in any of the RP experimental systems.

<sup>\*\* =</sup> Not measured.

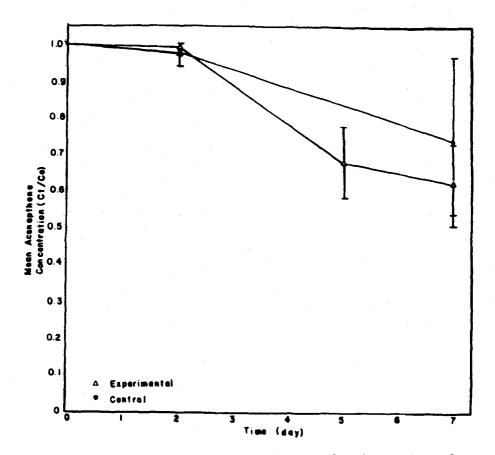
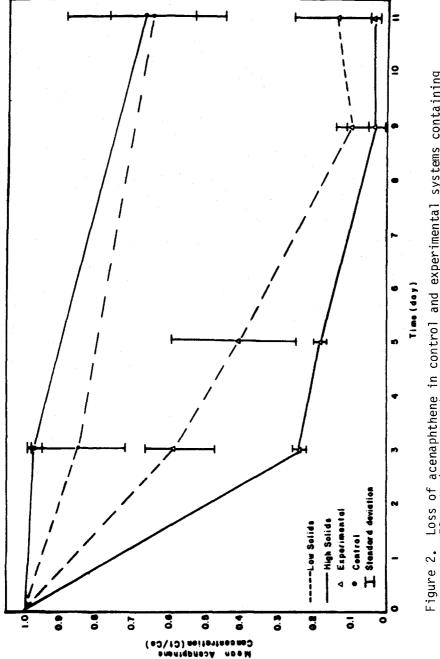


Figure 1. Loss of acenaphthene in control and experimental systems containing zero suspended solids.

Loss of acenaphthene in control and experimental systems containing RP sediments is illustrated in Figure 2. A 2 to 94 fold difference was observed between mean Ct/Co after 7 d $^{-1}$  in control and experimental systems with equal levels of PM sediments. Statistically significant differences were observed between rates of loss of acenaphthene in control and experimental systems containing suspended solids levels of 397 and 591 mgL $^{-1}$  PM sediments. Significant differences exist between rate of loss of acenaphthene in all PM experimental systems. Loss of acenaphthene in control and experimental systems. Loss of acenaphthene in control and experimental systems containing PM sediments is illustrated in Figure 3.

The fact that statistically significant differences were not found between rate of loss of acenaphthene in any of the RP experimental systems and significant differences were found between all PM



Loss of acenaphthene in control and experimental systems containing 52 mg/L (low solids) and 601 mg/L (high solids) RP sediments.

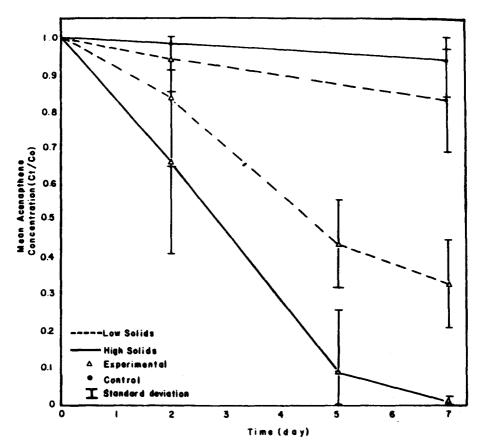


Figure 3. Loss of acenaphthene in control and experimental systems containing 83 mg/L (low solids) and 591 mg/L (high solids) PM sediments.

increased levels of systems indicates that experimental of loss of acenaphthene. sediments significantly increased rate Statistically significant differences were found between rates of loss of acenaphthene in RP and PM experimental exposures at the levels. highest suspended solids Acenaphthene would be expected to sorb to RP sediments to a larger extent than to PM sediments due to the higher organic carbon content of the former sediment type (Table 1). Sorption of acenaphthene to RP sediments have reduced the availability of acenaphthene to microbes indigenous to Roselawn Pond sediments. Table 3 lists kinetic data for loss of acenaphthene due to biological processes. This study physical-chemical characteristics demonstrated that suspended solids and/or microbes indigenous to different aquatic systems are factors influencing biotransformation of acenaphthene and possibly other polynuclear aromatic hydrocarbons.

Table 3. Kinetic data for experimental systems.

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Measured Solids (mg/l) <sup>1</sup>	r	T½.	К1			
0	0.89	24.75	0.028			
52 RP	0.95	3.52	0.197			
403 RP	0.88	4.03	0.172			
601 RP	0.91	2.23	0.311			
83 PM	0.92	4.91	0.141			
397 PM	0.91	1.20	0.577			
591 PM	0.83	0.83	0.831			

<sup>&</sup>lt;sup>1</sup>RP = Roselawn Pond Sediment; PM = Pat Mayse Lake Sediments.

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