

# USE OF A COMMERCIAL SURFACTANT FOR ENHANCED BIODEGRADATION OF ORGANIC WOOD-PRESERVATIVE CONTAMINATED PROCESSWATER.

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## ABSTRACT

A commercial surfactant (Brij 35) was studied in combination with and without a bacterial species known to break down pentachlorophenol (PCP) and polycyclic aromatic hydrocarbons (PAHs) associated with creosote. The effects of the surfactant, the bacterium, and the combination of the surfactant and bacterium, were evaluated in the biodegradation of process water highly contaminated with PAHs and PCP. Studies were conducted with 900 ml of process water in 1 liter amber jars. Nutrients along with the surfactant and/or bacterium were added to each jar and aeration was applied throughout the period of the study with an air sparger. Unconditioned controls and sterilized controls were also run. Samples were collected, extracted, and analyzed at 30 days and 60 days for PAHs and PCP, and also at day 90 for further PCP analysis. Total selected PAHs were significantly reduced in the surfactant and surfactant amended bacterial treatments. The results for PCP varied showing an increase during the study.

## INTRODUCTION

The wood-preserving industry has a variety of chemicals such as creosote and pentachlorophenol (PCP) in their treatment process and generate a large amount of process wastewater contaminated with various chemicals from several different sources including pressure treatment, steaming of wood, vapor drying and oil seasoning. The process wastewater produced today needs to be cleaned-up before the water is released. Also various sites exist throughout the country where ground water and soil have accumulated contaminants as a result of leaks, spills, rain water runoffs, and loose past regulations.

Bioremediation is well established as a less expensive technology in which microscopic organisms can detoxify and decompose organic wastes to meet clean-up standards for many contaminants (Thayer 1991), including pentachlorophenol (PCP) and polycyclic aromatic hydrocarbons (PAHs) from creosote (Stroo et al. 1989; Cerniglia 1992). By supplying the proper amount of nutrients, oxygen and additives needed for microbial cultures to grow, bioremediation of contaminated water and soil

may be accomplished in bioreactors and *in situ* (reviews, Maier 2000; Romantschuk et al. 2000). However, the limited availability of many environmental contaminants to microorganisms is a major factor that affects biodegradation. Surfactants can enhance the bioavailability of hydrophobic substances by increasing the solubility and surface area through the formation of micelles.

This study evaluated the effect of a commercial surfactant along with a PCP tolerant bacterium for their ability to enhance the biodegradation of wood-preserving process wastewater containing a high concentration of PAHs and PCP.

## MATERIALS AND METHODS

The process wastewater for this research was obtained from two wood treatment sites in Mississippi. One contained PCP and the other creosote. The two wastewaters were mixed vigorously with an industrial stainless steel blender to form a 1/1 (v/v) mixture. The pH was adjusted to 7. Three series of tests were conducted, one with a non-ionic surfactant Brij 35 (Acros Organics, Fairlawn, NJ) at ten times above critical micelle concentration (CMC = 74 mg / L), one with the addition of a bacterium known to tolerate the presence of PCP, and the other with the addition of both. Two sets of controls consisting of wastewater without the addition of surfactant or bacterium were also prepared. One set was sterilized by the addition of a 1.25% aqueous solution of sodium azide, while the other was not sterilized. The mixed wastewater (900 ml) was placed in 1 liter amber bottles with three replicates for each treatment. Nutrients were added as a 0.5% solution of Miracle-Gro (15:30:15). Oxygen was added continuously by sparging air, and bottles were stirred every 2-4 days. Every 30 days starting at day 0, 200 ml of the water was taken from each jar for continuous liquid-liquid extraction.

After continuous liquid-liquid extraction with dichloromethane (EPA Method 3520; U.S. EPA 1992), the sample was concentrated to 5 ml by Kuderna-Danish apparatus. Acidic and non-acidic (basic and neutral) portions of the sample were partitioned, the non-acidic portion was eluted through a Florisil (Floridin Co.) clean-up column (eluent dichloromethane) and analyzed

by gas chromatography for selected PAHs (Table 1) and PCP (EPA methods 8100 and 8040; U.S. EPA 1992). Bacteria were enumerated by the spread plate method on creosote and PCP selective media.

The data were statistically analyzed with a completely random design (CRD) on S.A.S. software (Release 6.12, Statistical Analysis System Institute, Cary, NC).

#### RESULTS AND DISCUSSION

The total concentration of selected PAHs dropped significantly at day 30 in all treatments including the control (Figure 1). Statistics revealed that the addition of the surfactant Brij 35 had a significant effect on the degradation of selected PAHs by day 60. The surfactant appears to have enhanced the bioavailability of PAHs in the water. The microbial counts (not listed in this paper), showed no correlation between bacterial population and the degradation rate of PAHs. Throughout the study, treatments amended with bacteria maintained moderate levels ( $10^6$  –  $10^8$  counts, colonies per mL) of creosote acclimated, PCP acclimated, creosote and PCP acclimated, and total bacteria. The control and surfactant treatments contained moderate levels of bacteria by day 15. The sterilized treatment contained  $10^5$  counts by day 30, and  $10^7$  counts by day 60. The addition of bacteria did not significantly affect the degradation of PAHs.

The results of gas chromatography analysis of PCP is depicted in Figure 2. Although an increase in the PCP concentrations was observed, statistical analysis of the results showed no significant increase nor decrease of PCP among treatments or over a time period of 90 days. This increase could be due to the initial effect of this surfactant on PCP absorbed to the suspended solids in process water.

In conclusion, the addition of surfactant Brij 35 enhanced the degradation of selected PAHs. However, no significant degradation of PCP was observed in this study. More detailed studies are needed to evaluate the effect of this surfactant on each specific PAHs. Further studies are also needed to determine how quickly surfactants like Brij 35 could desorb compounds like PCP from suspended solids in process water.

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**Table 1. List of selected polycyclic aromatic hydrocarbons (PAHs) analyzed by gas chromatography in this study.**

naphthalene	acenaphthene	1,2-benzanthracene
1-methylnaphthalene	acenaphthylene	chrysene
2-methylnaphthalene	fluorene	pyrene
biphenyl	carbazole	benzo[a]pyrene
anthracene	dibenzofuran	benzo[ghi]perylene
phenanthrene	fluoranthene	

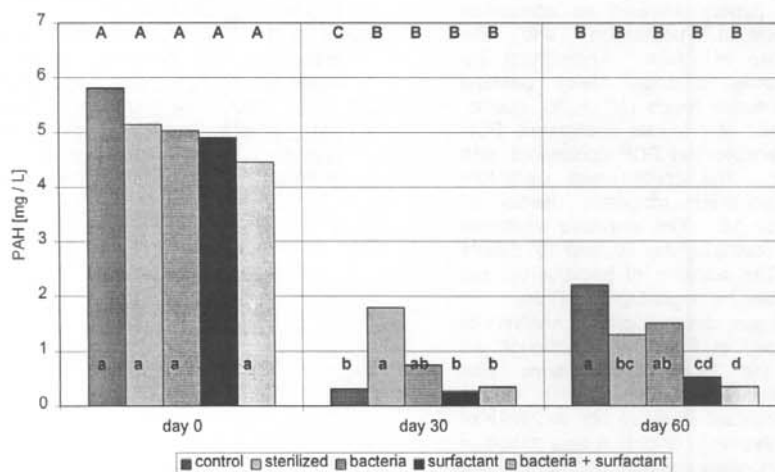


Figure 1. Gas chromatographic analysis of the degradation of total selected polycyclic aromatic hydrocarbons (PAHs) in process water. Each column represents an average of three replicates. Different lower case letters on columns indicate significant differences among treatments at each sampling date. Different capital letters above columns indicate significant differences over time for each treatment.

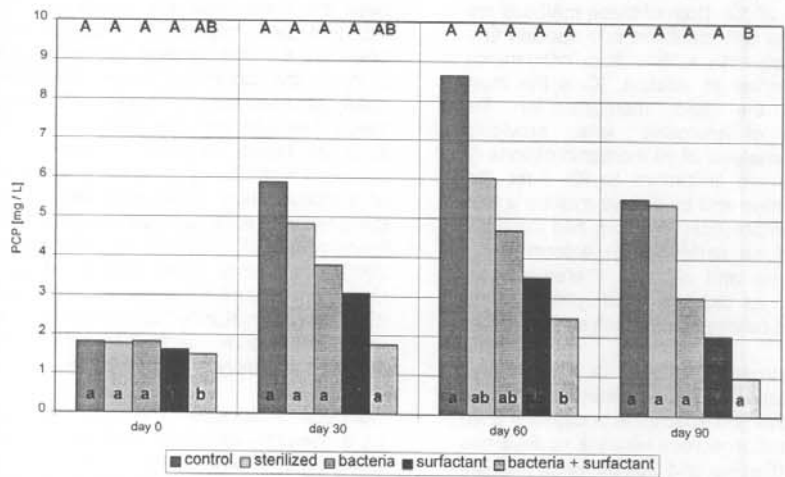


Figure 2. Gas chromatographic analysis of the degradation of pentachlorophenol (PCP) in process water. Each column represents an average of three replicates. Different lower case letters on columns indicate significant differences among treatments at each sampling date. Different capital letters above columns indicate significant differences over time for each treatment.

