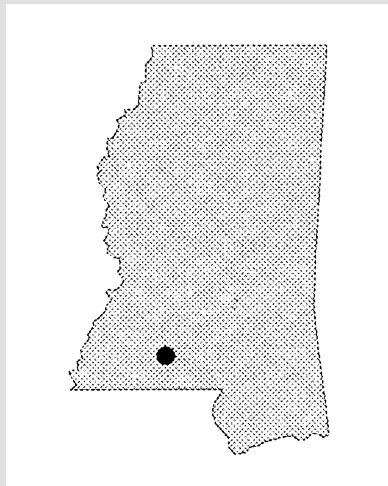




## SITE FACTS



**Location:** Brookhaven,  
Mississippi

**Laboratories/Agencies:** U.S.  
EPA National Risk Management  
Research Laboratory (NRMRL),  
U.S. Department of Agriculture  
Forest Products Laboratory  
(FPL), Superfund Innovative  
Technology Evaluation (SITE)  
Program, U.S. EPA Region 4

**Media and Contaminants:**  
Pentachlorophenol (PCP) and  
creosote in soil

**Treatment:** Lignin-degrading  
fungi

**Date of Initiative Selection:**  
Spring 1991

**Objective:** To evaluate the  
effectiveness of lignin-degrading  
fungi treatment for wood  
preserving wastes

### **Bioremediation Field Initiative**

**Contacts:** John Glaser and  
Richard Brenner, U.S. EPA  
NRMRL, 26 West Martin Luther  
King Drive, Cincinnati, OH 45268

**Regional Contact:**  
De'Lyntoneus Moore, U.S. EPA  
Region 4, Waste Management  
Division, 345 Courtland Street,  
Atlanta, GA 30365

# Bioremediation Field Initiative Site Profile: Escambia Wood Preserving Site

## Background

The Escambia Wood Preserving Site located in Brookhaven, Mississippi, is a former wood preserving facility that used pentachlorophenol (PCP) and creosote to treat wooden poles. Located on the site are two pressure treatment cylinders, a wastewater treatment system, five bulk product storage tanks, seven condenser ponds (including a 3,000,000-gal., unlined primary surface impoundment), and excavated soil materials containing process chemicals from past spills. In April 1991, U.S. EPA Region 4 initiated a removal action to eliminate all sources of potential releases to the environment.

In the fall of 1991, PCP-contaminated soil from previous spill events was used to evaluate multiple soil treatment regimes involving three different fungal strains during an 8-week treatability study. A follow-on field-scale demonstration study using one specific strain of lignin-degrading fungi was conducted over a 5-month period (June to November 1992). Both studies were carried out by the U.S. EPA Risk Reduction Engineering Laboratory (RREL) (now the National Risk Management Research Laboratory [NRMRL]) and the U.S. Department of Agriculture Forest Products Laboratory (FPL) under the Superfund Innovative Technology Evaluation (SITE) Program with support from the Bioremediation Field Initiative.

## Site Characterization

In preparation for conducting a treatability study, candidate contaminated source soils from the site were sampled at the surface in June 1991. Composite soil samples were analyzed for PCP and other volatile and semi-volatile organics. The composite samples contained elevated concentrations of 44 organic compounds, 12 of which are hazardous constituents of K001 waste. PCP concentrations were found to range from 25 to 342 mg/kg, with an average of 143 mg/kg. Contaminant concentrations varied greatly within the sampled soil. Subsequent excavation of these soils to provide sufficient quantities of material for the treatability and demonstration studies showed increasing concentrations of PCP with depth, eventually encountering concentrations in excess of 5,000 mg/kg.

## Treatability Study

The treatability study compared 10 treatments, combining three fungal species, three inoculum loading levels, and the appropriate controls. The experimental method combined a randomized complete block (RCB) design without replication and a balanced incomplete block (BIB) design with treatment replicated four times. Eleven 10-ft by 10-ft plots, each holding about 4 tons of soil, were constructed. In the RCB design, six of the plots each received a separate treatment. In the BIB design, each of the five remaining plots was divided by interior borders into four 2.5-ft. by 2.5-ft



split plots. These plots were used to evaluate one of the treatments from the RCB design and four additional treatments.

Excavated soil was mechanically sieved to pass through a 1-in. screen, mixed, and placed in the plots to a depth of 10-in. The plots were then inoculated with the fungi. After inoculation, each plot periodically was irrigated and tilled with a garden rototiller. Wood chips were added to each plot to provide a substrate to sustain growth of the fungi. Figure 1 is a schematic of the soil preparation, showing the treatment plots. Treatment with *Phanerochaete chrysosporium* resulted in an average 69 percent reduction in PCP for initial concentrations in excess of 1,000 mg/kg, while an average reduction of 89 percent was achieved with *Phanerochaete sordida* with initial PCP concentrations of 600-700 mg/kg.

## Demonstration Study

A field-scale demonstration study was conducted in 1992 using the lignin-degrading fungus *P. sordida* to treat PCP and 13 polycyclic aromatic hydrocarbons (PAHs) found in the contaminated soil. *P. sordida* was selected based on the results of the treatability study and its natural occurrence in soil. The soil excavated for this study had much higher PCP concentrations (up to 5,200 mg/kg) than that used in the treatability study, although the average concentration was decreased to approximately 1,000 mg/kg by dilution with clean soil. The contaminated soil was placed in a 70-ft by 100-ft soil treatment bed and two 25-ft by 50-ft control beds. The treatment bed was inoculated with 10 percent dry (w/w) fungal hyphae and inoculum substrate/soil. No amendments were added to one of the control beds, while the other was formed by blending in 10 percent dry (w/w) sterile inoculum.

Depletion of targeted compounds in the treatment bed and the two control beds was measured over a 20-week period from June to November. A 64 percent reduction in PCP concentration was achieved in the fungal treatment bed compared to 26 percent in the sterile inoculum-amended control bed and 18 percent in the non-amended control bed. Substantial reductions in the concentrations of 3- and 4-ring PAHs were noted in all three beds, although depletion of 4-ring PAHs was greater in the treatment bed than in the control beds. The 5- and

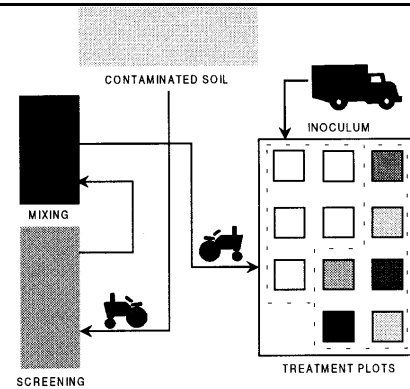


Figure 1. Schematic of soil preparation, from excavation to screening, mixing, placement in treatment plots, and inoculation with fungi.

6-ring PAHs were not significantly diminished in either the treatment or the control beds. The extent of treatment was controlled by two factors: low biological activity of the fungal inoculum as determined by ergosterol measurements at the beginning of the study and unfavorable weather conditions that severely curtailed the desired tilling schedule for the soil beds.

## Status

Although the treatment efficiencies achieved in the 5-month field-scale demonstration study at Brookhaven were somewhat disappointing after the highly promising treatability study, the demonstrated ability of lignin-degrading fungi to remove significant quantities of PCP at an initial concentration of approximately 1,000 mg/kg is considered encouraging. This concentration is roughly 10 times greater than concentrations typically attempted with bacterial-based treatments. The excessive rainfall encountered at Brookhaven, which frequently interfered with regularly scheduled tilling, may have resulted in oxygen depletion in the treatment bed, leading to loss of fungal mass and activity. This environmental consequence suggests that future applications of fungal treatment technology may need to avoid soil bed configurations to prevent weather-related constraints. Current costs of fungal treatment operations are estimated at \$150-200/ton of soil. Reductions in inoculum mass requirements and costs are anticipated as new inoculum formulations and application techniques are developed.

**The Bioremediation Field Initiative** was established in 1990 to expand the nation's field experience in bioremediation technologies. The Initiative's objectives are to more fully document the performance of full-scale applications of bioremediation; provide technical assistance to regional and state site managers; and provide information on treatability studies, design, and operation of bioremediation projects. The Initiative currently is performing field evaluations of bioremediation at eight other hazardous waste sites: Libby Ground Water Superfund site, Libby, MT; Park City Pipeline, Park City, KS; Bendix Corporation/Allied Automotive Superfund site, St. Joseph, MI; West KL Avenue Landfill Superfund site, Kalamazoo, MI; Eielson Air Force Base Superfund site, Fairbanks, AK; Hill Air Force Base Superfund site, Salt Lake City, UT; Reilly Tar and Chemical Corporation Superfund site, St. Louis Park, MN; and Public Service Company, Denver, CO. To obtain profiles on these additional sites or to be added to the Initiative's mailing list, call 513-569-7562. For further information on the Bioremediation Field Initiative, contact Fran Kremer, Coordinator, Bioremediation Field Initiative, U.S. EPA, Office of Research and Development, 26 West Martin Luther King Drive, Cincinnati, OH 45268; or Michael Forlini, U.S. EPA, Technology Innovation Office, Office of Solid Waste and Emergency Response, 401 M Street, SW., Washington, DC 20460.