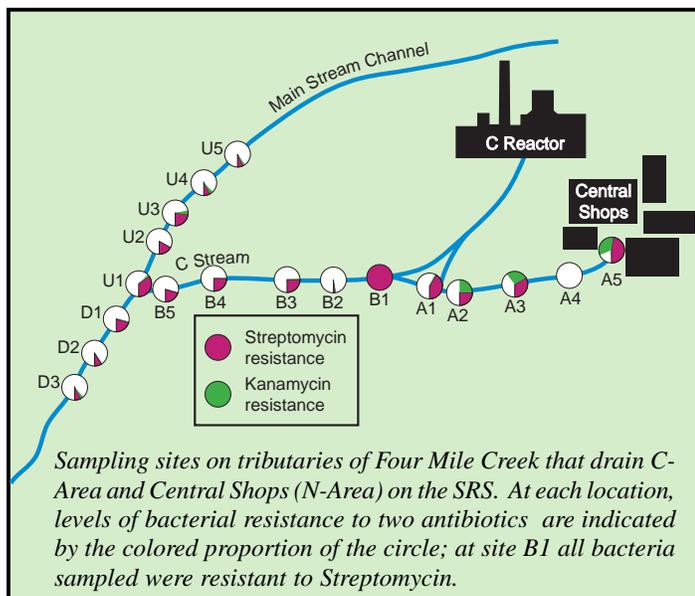
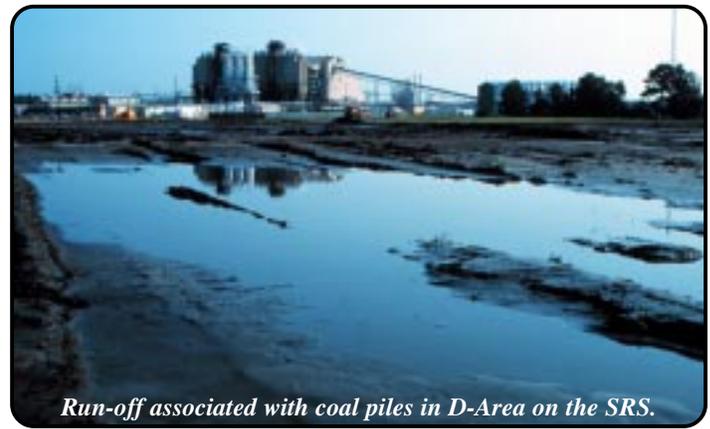


primary focus of many bioremediation studies. While some bacteria respond to perturbations with increased growth rates, it is not clear what effect various forms of pollution have on bacterial species or genetic diversity in the native communities. For example, one result of metal pollution is an increase in the numbers and kinds of metal-resistant bacteria. The genes that code for metal resistance are often carried on plasmids, or small mobile pieces of DNA. Coincidentally, these same plasmids often carry genes that confer antibiotic resistance. In a survey of bacterial assemblages collected from Four Mile Creek on the SRS, we found that:

- highest levels of antibiotic resistance were found in bacteria in a tributary stream that drains Central Shops and C-Reactor,
- there may be significant industrial contamination in this tributary but not in the main stream channel, and
- it appears that antibiotic resistance may be a good indicator of level of contamination and thus need for cleanup.



Given the large areas of the earth that have industrial pollution, these results may be even more significant. Because antibiotic resistance is carried on mobile DNA elements, it can be distributed easily to unrelated bacteria, including pathogens. Our current research is investigating whether antibiotic resistant bacteria can escape from contaminated streams into the atmosphere. If so, we are trying to determine how far they are transported into the atmosphere (i.e., meters, kilometers, worldwide). This is the first study that is attempting to link the indirect effect of industrial pollution to the resurgence of antibiotic resistance in human pathogens.

Ecology of bacteria associated with coal pile run-off

The storage of sulfur-rich coal and the combustion products of coal represents a pollution source that has severely impacted numerous ecosystems. Specifically, the exposure of coal deposits to oxygen and water results in the conversion of pyrite (FeS_2) to sulfuric acid. The resulting acidic leachate is enriched

with salts and heavy metals, forming a type of pollution referred to as acid mine drainage (AMD). Despite the fundamental role of bacteria as both causative agents and as potential bioremediators of AMD, their ecology in these systems has not been studied in a comprehensive manner, due primarily to an inability to culture the majority of environmental bacteria.

D-Area on the SRS has a 20-year-old exposed reject coal pile. Acidic, metal-rich leachate from this pile has contaminated the sediments of an adjacent forested wetland, causing vegetation die-off. Historically, because of the need to culture or grow bacteria, it has been extremely difficult to monitor changes in the total bacterial community since less than 1% of the bacteria in a sample can be cultured. However, recent advances in molecular biology allow scientists to track, under field conditions, various bacterial genes or gene sequences without the requirement of culturing the bacteria. Our research project is using some of these molecular biological tools to determine the bacterial community composition within a contamination gradient. This information will be used to determine if and how bacterial community composition and diversity has changed in response to the acidic, metal-rich coal leachate. Additionally, this experiment could provide data to test the hypothesis that as yet uncultured bacteria, in addition to the well-studied bacteria *Thiobacillus ferrooxidans* and *Leptospirillum ferrooxidans*, could be causative agents of AMD.

Cleanup of contaminated sites within the DOE complex is a major budgetary concern. Bacteria have been shown to be effective indicators and processors of contamination. SREL studies will provide information on the basic ecology of bacteria that will allow managers to make informed decisions on the feasibility of technology transfer to other sites. In addition, these studies may demonstrate a potential new dimension to risk assessment, i.e., the risk of the dissemination of antibiotic resistance and its subsequent effects on human health.

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