



**US Army Corps
of Engineers.**
Construction Engineering
Research Laboratory

Fact Sheet

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PATHWAYS AND CONTROLLING FACTORS IN THE BIODEGRADATION OF ENERGETIC WASTES

The Problem

Wastewaters containing explosives are produced at Army installations carrying out manufacturing, and loading, assembly, and packing operations. Approximately 35 million pounds of pink water containing trinitrotoluene (TNT), 2,4-dinitrotoluene (DNT), and cyclonitramines (RDX and HMX) are produced annually at IOC installations. These wastewaters must be treated to comply with environmental laws and regulations. Compounds found in pink water are resistant to aerobic biodegradation and are toxic, therefore Army installations are prohibited from discharging wastewater into local municipal wastewater treatment plants. Compounds found in pink water are resistant to aerobic biodegradation and are toxic, therefore Army installations are prohibited from discharging pink water into local municipal wastewater treatment plants. Granular activated carbon (GAC) adsorption is currently the standard technology for treating and maintaining discharges within limits. GAC is effective, but expensive, and the explosive-laden GAC must periodically be transported off-site for regeneration or destruction by incineration.

The Technology

Biodegradation is the only means (other than incineration) by which toxic compounds can be transformed into harmless water and carbon dioxide (and/or methane). Explosive compounds tend to be resistant to degradation under aerobic conditions, but are readily transformed by bacteria under anaerobic conditions. Although such studies demonstrate the potential usefulness for treating explosive contaminated wastewaters anaerobically, much work remains to be done. The environmental and physiological factors controlling the biodegradation of most nitroaromatic and explosive compounds are not well understood, nor are the biodegradation pathways well established. As more is learned about the factors limiting their degradation, strategies can be developed to overcome them, improving the performance and efficiency of biological treatment processes, decreasing treatment costs even more. As research studies move from studying microorganisms to the enzymes degrading explosives, enzyme-based reactors can be developed. These overcome many of the disadvantages and limitations of bioreactors using whole cells.

Benefits/Savings

Developing efficient biological treatment technologies will allow the Army to cost effectively treat wastewater, maintain effluent quality, and help meet new environmental regulations.

Status

This is the last year of a five-year basic research project studying the factors limiting the transformation of explosive compounds. Significant progress has been made in understanding how bacteria degrade energetic compounds anaerobically. Most of our studies were conducted with a culture isolated from the Holston Army Ammunition wastewater treatment plant. The culture readily degrades RDX, HMX, and TNT when ethanol is added as a cosubstrate. In the absence of an explosive compound, ethanol is mineralized to CH₄ and CO₂, but in its presence, methane production is inhibited. Initially we thought this was due to the toxicity of the energetic compound to the methanogenic bacteria. Later studies demonstrated, however, that ethanol was degraded and acetate accumulated. This only occurs if H₂, produced during the transformation of ethanol to acetate, is removed from the system. Methanogenic bacteria usually remove H₂ and produce methane, but in the presence of an explosive compound there was none. Interestingly, the culture contained relatively high numbers of acetogenic bacteria, known for their ability to oxidize H₂. Their presence in the culture and the absence of methane production from ethanol when an explosive is added to the culture indicates they may be responsible for H₂ removal as well as degrading the explosive compounds. Future work will concentrate on isolating the acetogenic bacteria from the culture and determining their ability to degrade the various explosives.

Understanding how explosives are degraded helps us develop strategies to overcome the factors limiting their degradation. Studying the microbial ecology of our culture is essential to make effective control of the startup and operations of anaerobic bioreactors possible. Furthermore, these results can be extrapolated to the field. As our understanding increases, predictions can be made regarding explosive degradation *in situ* and strategies developed to enhance the biodegradation of explosives in contaminated groundwater. The addition of methanogenic cosubstrates appears to be a strategy for accelerating the *in situ* biodegradation of these compounds.

Point of Contact

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