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Monitored Natural Attenuation of Explosives

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Explosives are subject to several attenuation processes that potentially reduce concentrations in groundwater over time. Some of these processes are well defined, while others are poorly understood. The objective of the project was to optimize data collection and processing procedures for evaluation and implementation of monitored natural attenuation of explosives. After conducting experiments to optimize data quality, a protocol was established for quarterly monitoring of thirty wells over a 2-year period at a former waste disposal site. Microbial biomarkers and stable isotopes of nitrogen and carbon were explored as additional approaches to tracking attenuation processes. The project included a cone penetrometry sampling event to characterize site lithology and to obtain sample material for biomarker studies. A three-dimensional groundwater model was applied to conceptualize and predict future behavior of the contaminant plume. The groundwater monitoring data demonstrated declining concentrations of explosives over the 2 years. Biomarker data showed the potential for microbial degradation and provided an estimate of the degradation rate. Measuring stable isotopic fractions of nitrogen in TNT was a promising method of monitoring TNT attenuation. Overall, results of the demonstration suggest that monitored natural attenuation is a viable option that should be among the options considered for remediation of explosives-contaminated sites.

KEY WORDS: *TNT, RDX, groundwater monitoring, microbial biomarkers, stable isotopes, groundwater modeling.*

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Background

Explosives contamination of soil and groundwater has resulted from manufacturing and load-assemble-package (LAP) processes conducted in the U.S. primarily during and before World War II. Principal explosives waste products were 2,4,6-trinitrotoluene (TNT) and hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX). Waste disposal practices were governed more by convenience and explosives safety concerns than by environmental concerns because environmental awareness in the U.S. was poorly developed. Wastewater was often discharged into sumps, runoff, and/or percolation ditches, and lagoons, resulting in the contamination of soil and groundwater.

Several processes have been explored for remediation of explosives-contaminated groundwater. The most widely used has been adsorption of explosives by granular activated charcoal after pumping of groundwater to the surface. When charcoal columns are placed in series, this process is very cost effective (about $1 [dollar] per thousand gallons) for explosives concentrations less than one part per million (Marks *et al.*, 1995). For concentrations above 1 ppm, pretreatment with an ultraviolet oxidation systems followed by carbon adsorption columns (UV/Ox) may be more cost effective. Phytoremediation in flow-through wetlands constructed of gravel beds effectively removes TNT, its related products, and RDX (Best *et al.*, 1999). However, as with UV/Ox, phytoremediation typically requires pumping of ground water to the surface, which can increase remediation costs. *In situ* processes, such as construction of biobarriers or biologically active zones, have been problematic due to the recalcitrance of explosives to microbial mineralization and difficulties in maintaining flow and microbial activity in various subsurface media. Monitored natural attenuation may be an attractive remedial alternative for explosives-contaminated sites that meet well-defined selection criteria and for which acceptable risk levels can be demonstrated.

The Louisiana Army Ammunition Plant (LAAP) was selected for a 2-year project designed to demonstrate monitored natural attenuation (MNA) as a remedial alternative for explosives in groundwater (Figure 1). The LAAP was selected because the source of contamination had already been remediated, a large body of contaminant data was available, and the site was well characterized with approximately 50 functional groundwater monitoring wells in place. The demonstration was conducted at Area P where wastes from LAP of munitions had been disposed into 16 unlined lagoons (Figure 1). The lagoons had been pumped out and the sediment excavated and incinerated in the late 1980s. Soil was excavated until a total field-determined concentration of less than 100 mg explosives per kg of soil was achieved (SAIC, 1994). The area was capped with compacted clay overlaid with 4 in of topsoil with a slope of at least 1% to facilitate drainage. The clay was compacted to at least 90% of the standard proctor density for the clay used. At the initiation of the MNA demonstration, the area was managed for low-growing natural vegetation consisting primarily of invasive grasses and low herbs.

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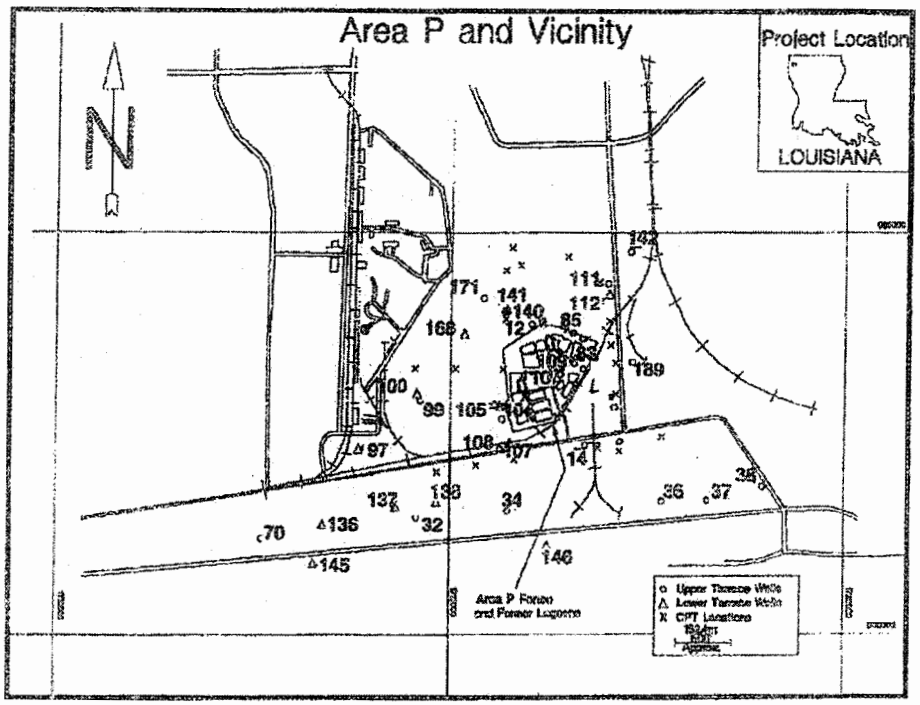

FIGURE 1

Locations of groundwater monitoring wells and cone penetrometry (CPT) sampling sites at Area P, Louisiana Army Ammunition Plant.

The near surface geology at LAAP consists of Pleistocene, terraced fluvial deposits (basal gravels fining upward to clays) unconformably overlying Eocene, nonmarine, massive sands, silty sands, silty clays, and occasional lignitic beds. An effectively impermeable boundary, the Cane River Formation, lies below the fluvial deposits. Groundwater plumes containing TNT, RDX, and 1,3,5-trinitrobenzene (TNB, a photodecomposition product of TNT) had been detected beneath Area P (SAIC, 1994). Concentrations in the upper terrace aquifer were lower in 1994 than in 1990, suggesting an improvement in the groundwater quality since the removal of the lagoons. The groundwater contaminant plumes had not advanced very far laterally, suggesting very slow transport and the potential for attenuation (Figure 2, conceptualization of the TNT plume at the inception of the demonstration).

Attenuation Processes

Transformation, immobilization, and biodegradation are potential attenuation processes for explosives. Transformation occurs either biotically (Kaplan, 1993;

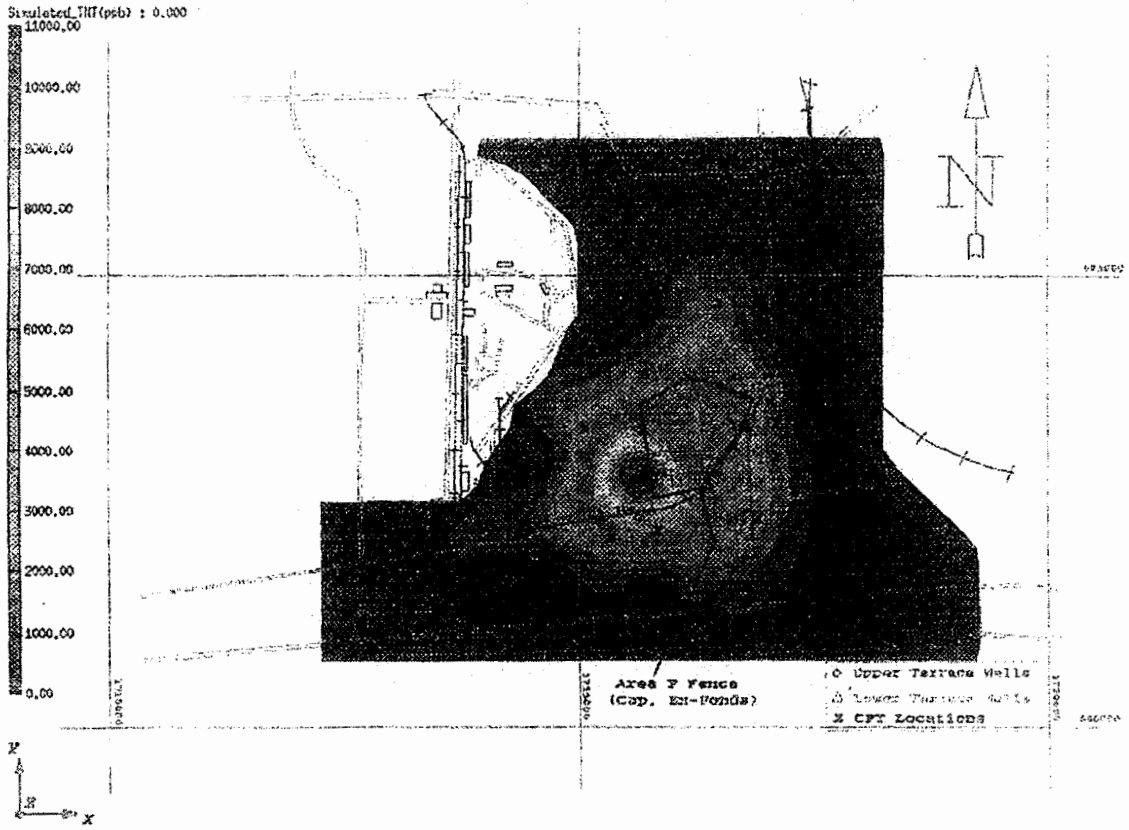

FIGURE 2

Conceptualization of the lateral extent of the TNT groundwater plume at the initiation of the study. Units on vertical scale are ppb.

Kaplan *et al.*, 1985; Kaplan and Kaplan, 1982; McCormick *et al.*, 1981) or abiotically (Brannon *et al.*, 1998; Pennington and Patrick, 1990; Sitzman, 1974) by the reduction of the nitro moities of TNT to amino groups (Figure 3) and the nitro

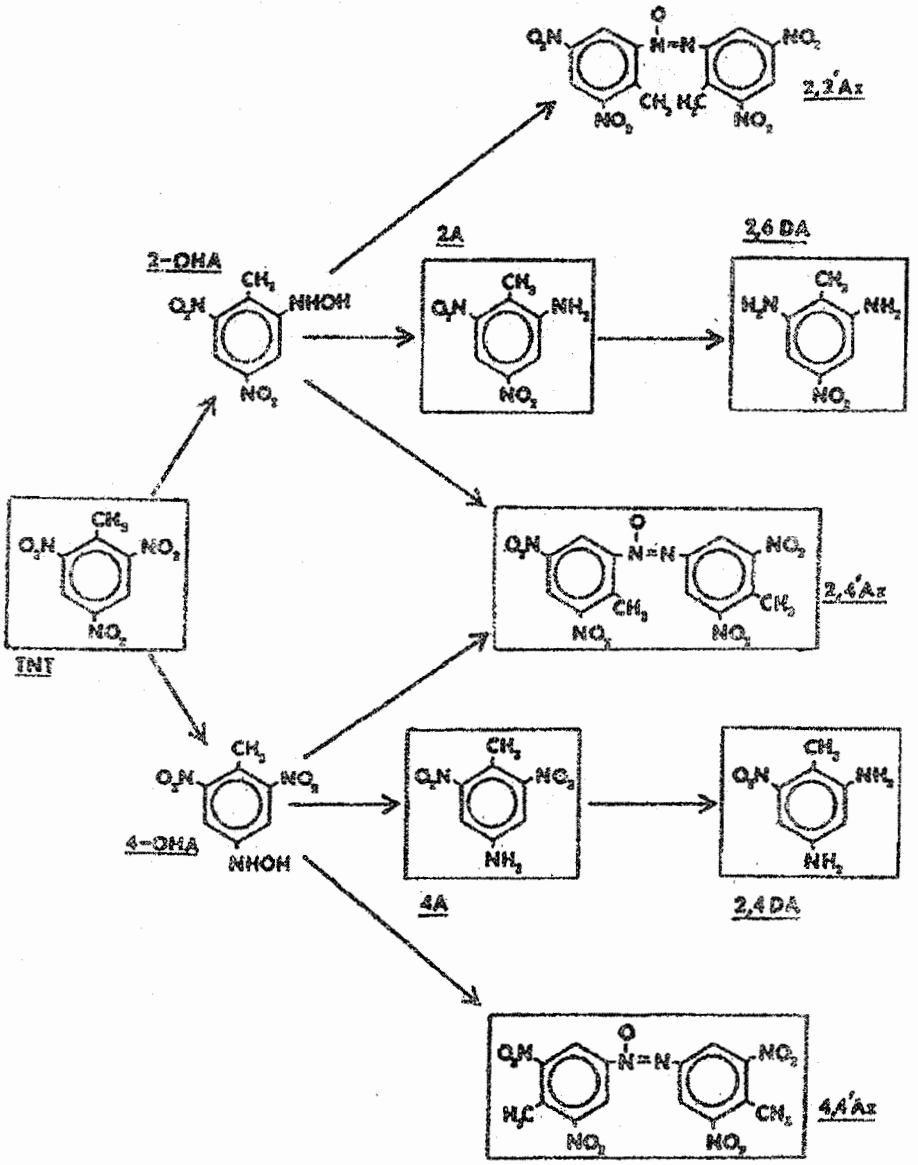

FIGURE 3

Principal transformation pathway for TNT (Kaplan and Kaplan, 1982). Abbreviations for compounds are as follows: 2-OHA, 2-hydroxylamino-4,6-dinitrotoluene; 4-OHA, 4-hydroxylamino-2,6-dinitrotoluene; 2A, 2-amino-4,6-dinitrotoluene; 4A, 4-amino-2,6-dinitrotoluene; 2,2'Az, 4,4'6,6'-tetranitro-2,2'-azoxytoluene; 2,6DA, 2,6-diamino-4-nitrotoluene; 2,4'Az, 4,2',6,6'-tetranitro-2,4'-azoxyutoluene; 2,4DA, 2,4-diamino-6-nitrotoluene; 2,2'6,6'-tetranitro-4,4'-azoxytoluene. Boxed compounds were identified in tests; unboxed compounds were proposed.

FIGURE 2

Conceptualization of the lateral extent of the TNT groundwater plume at the initiation of the study. Units on vertical scale are ppb.

moities of RDX to nitroso groups (Figure 4). Some of these transformation products are of environmental concern and must be removed along with the parent compound to effect adequate site cleanup (Honeycutt *et al.*, 1996; Jarvis *et al.*, 1998; Tan *et al.*, 1992; Won *et al.*, 1976). The hydroxylamino, monoamino, diamino, and azoxy transformation products of TNT were first identified in a simulated compost system (Figure 3, Kaplan and Kaplan, 1982). The hydroxylamino products are rarely observed because of their instability. The diamino and azoxy products are also rarely observed in natural systems. However, the monoamino transformation products are common where TNT is found. Detection of transformation products of RDX is uncommon.

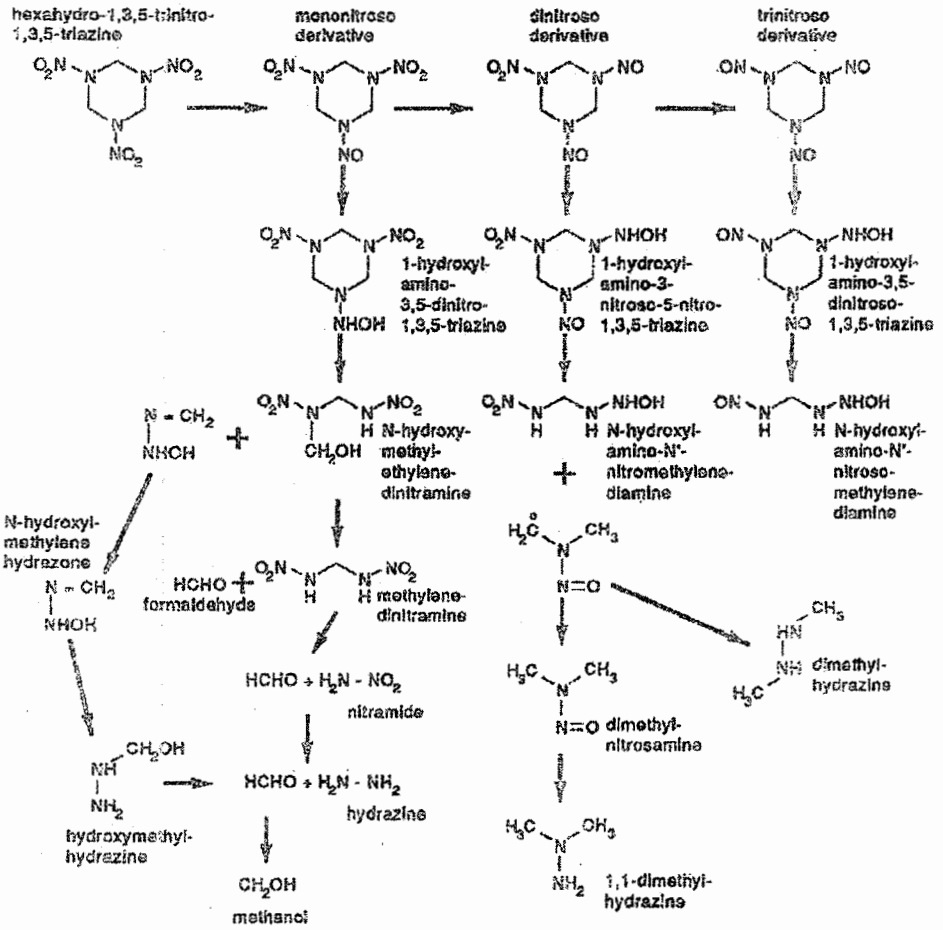

FIGURE 4

Anaerobic biomineralization pathway for RDX (McCormick, Feeherry, and Levinson, 1976; Kaplan, 1993).

The amino transformation products of TNT are potentially reactive with functional groups on organic matter in the soil to produce complex products that are no longer mobile. Evidence for these immobilization products has been observed in the high organic carbon matrix of compost (Pennington *et al.*, 1995; Caton *et al.*, 1994; Kaplan and Kaplan, 1982) and in soil organic matter (Price *et al.*, 1997). Recent studies using nuclear magnetic resonance techniques and TNT transformation products labeled with stable isotopes of nitrogen demonstrate the covalent bonding of these reactions (Thorn, 1998).

Many investigators have examined microbial degradation of nitroaromatic compounds using selected microbial communities (Osmon and Klausmeier, 1972; Carpenter *et al.*, 1978; Kaplan and Kaplan, 1982; Hallas and Alexander, 1983; Fernando *et al.*, 1990; Valli *et al.*, 1992; Funk *et al.*, 1993). However, surprisingly little is known about degradation of nitroaromatics in situ. A mineralization pathway for TNT has been proposed (Duque *et al.*, 1993; Preuss *et al.*, 1993); however, TNT is only slowly mineralized in soils and groundwater (Pennington *et al.*, 1999). RDX is relatively unaffected by transformation compared to TNT and is more readily mineralized, especially under anaerobic conditions (McCormick *et al.*, 1985). An anaerobic mineralization pathway for RDX has been proposed (Figure 4, Kaplan, 1993).

Approach at LAAP

Monitored natural attenuation relies heavily on development of a defensible "weight-of-evidence" that natural processes will be protective of potential receptors. To that end, the U.S. Environmental Protection Agency identifies the following three "tiers of site-specific information, or 'lines of evidence' " in OSWER directive 9200.4-17P (USEPA, 1999):

1. Historical groundwater and/or soil chemistry data that demonstrate a clear and meaningful trend of decreasing contaminant mass and/or concentration over time at appropriate monitoring or sampling points.

2. Hydrogeologic and geochemical data that can be used to demonstrate indirectly the type(s) of natural attenuation processes active at the site, and the rate at which such processes will reduce contaminant concentrations to required levels.

3. Data from field or microcosm studies (conducted in or with actual contaminated site media) which directly demonstrate the occurrence of a particular natural attenuation process at the site and its ability to degrade the contaminants of concern (typically used to demonstrate biological degradation processes only).

In order to address each of the above, the demonstration project at LAAP included the following tasks:

1. Review of pre-project groundwater data for trends in contaminant concentration over time
2. Development and implementation of a 2-year groundwater monitoring plan to define trends in contaminant concentration
3. Collection of aquifer soils and water to refine site characterization (e.g., lithology, hydrogeology, resistivity, contaminant concentrations) and to provide subsurface sample material for biomarker studies
4. Measurements of geochemical capacity of the site for explosives attenuation in laboratory microcosms
5. Investigations of three techniques for evaluating microbial activity at the site
6. Investigation of the feasibility of using stable isotopes of carbon and nitrogen to track attenuation
7. Groundwater modeling for conceptualization of the plume and for predicting future contaminant behavior

Analysis of Pre-Project Contaminant Data

Historical data for Area P were available for the time period 1980 to 1995. Data collected prior to 1986 was not used because it lacked precision relative to later data, and because methods descriptions were unavailable. Data having detection limits equal to or greater than 4.9 $\mu g\ L^{-1}$ were considered imprecise and were also excluded. All remaining data consisting of less than detection limit values were set equal to the detection limit for the statistical analyses. Data for Upper Terrace aquifer (32 wells) and Lower Terrace aquifer (29 wells) were analyzed separately. Mean yearly concentrations for each explosive from each well were used to minimize seasonal influences. Wells were analyzed in two groups because of substantial data gaps in the time series data. The groups were wells exhibiting concentrations greater than detection limit at some point during the time period, but always $<10\ \mu g\ L^{-1}$ (outer zone) and wells exhibiting concentrations greater than $10\ \mu g\ L^{-1}$ (inner zone).

For each explosive, terrace, and zone, the normality of untransformed and log-transformed concentration data were tested using Shapiro-Wilk's Test. Normality was rejected if the W statistic was <0.90. For most explosives, log transformation improved normality. Least-squares regression of contaminant concentration on year was performed for each explosive, terrace, and zone, usually with log-

transformed data. A significant regression F statistic ($P < 0.05$) indicated an increasing trend in concentration over time when the slope was positive, and a decreasing trend when the slope was negative. Lack-of-fit analyses were performed for each regression model. Fit was rejected if P were less than 0.05 for the lack-of-fit F statistic, regardless of whether the regression slope was significant. If normality could not be satisfied using either untransformed or log-transformed data, a nonparametric test for trend was performed by calculating Spearman's rank correlation statistic (ρ) between contaminant concentrations and sample year. A significant positive Δ indicated an increasing trend; a negative ρ indicated a decreasing trend.

Data were extremely variable exhibiting a range of up to four orders of magnitude in a sample year. Fewer trends were evident in the upper than in the lower terrace (Table 1). Decreasing trends were observed for TNT, TNB, and octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine (HMX) in the outer zone of the lower terrace aquifer. However, increasing trends were observed in TNT, TNB, 1,3-dinitrobenzene (DNB), 2,4-dinitrotoluene (2,4DNT), and HMX in the inner zone of the lower terrace aquifer. The only trends observed in the upper terrace were decreases in TNB, and DNB in the outer zone, decreases in HMX and nitrobenzene (NB) in the inner zone, and an increase in 2,4DNT in the inner zone. No significant trends over time were observed for RDX, *N*-methyl-*N*,2,4,6-tetranitrobenzenamine (tetryl), or 2,6-dintrotoluene (2,6DNT).

Cone Penetrometry Sampling

Soil samples were collected from 24 locations along 8 transects using cone penetrometry (CPT) (Figure 1). Generally, the penetrations were about 15 m (50 ft) deep and reached the confining layer. Locations were determined to meet the following sampling objectives: (1) to refine the vertical and lateral definition of the contamination, (2) to refine site lithology and hydrogeology, and (3) to provide subsurface soil samples for other tests. Sampling on the Area P cap was not permitted due to concern that the integrity of the cap would be compromised. TNT and RDX concentrations from groundwater sampling prior to the date of the CPT event were contoured to identify "hot spots" prior to establishing transects. The first punch on a given transect was used to stratify the site by measuring resistivity to penetration. The strata were defined in terms of lithology. Subsequent punches were used to collect soil samples with a presterilized split spoon. Typically, at least three depths, surface, mid-depth, and just above the confining layer, were sampled. Sampling along each transect was restricted to one or two locations before proceeding to the next transect. This allowed time to send the samples to the laboratory and obtain explosives concentration data on which to base selection of the next location along that transect. Vertical profiles of soil samples were collected at five locations. Depending on the depth of the CPT hole, from 6 to 14 samples were

Table 1. Summary of Trends in Explosives Concentrations Over Time in the Area P Historical Data (1986-1995)[1]

Explosive[2]	Upper Terrace						Lower Terrace					
	Inner Zone			Outer Zone			Inner Zone			Outer Zone		
	N	Model	Trend	N	Model	Trend	N	Model	Trend	N	Model	Trend
TNT	28	Log-linear F=0.36 P=0.55	None	5	Log-linear F=3.59 P=0.15	None	23	Log-linear F=6.36 P=0.02	Increasing	17	Nonparametric Δ=-0.58 P=0.01	Decreasing[3]
RDX	36	Log-linear F=0.13 P=0.72	None	8	Log-linear F=2.04 P=0.88	None	44	Log-linear F=2.81 P=0.10	None	17	Log-linear F=3.18 P=0.09	None
TNB	26	Log-linear F=1.04 P=0.32	None	36	Log-quadratic F=4.64 P=0.02	Decreasing	20	Log-linear F=13.88 P=0.002	Increasing	35	Log-linear F=6.17 P=0.02	Decreasing

[1]Most wells were sampled during 1 or 2 years between 1986 and 1995. No wells were sampled in 1992. Sixty-two wells were sampled in 1990. Because of the substantial data gaps, wells were grouped into two zones, a zone of higher concentration ($\geq 10 \mu g\ L^{-1}$) and a zone of lower concentration ($< 10 \mu g\ L^{-1}$) in each terrace.

[2]Other explosives analytes for which results are not shown include the following: 1,3-dinitrobenzene, 2,4-dinitrotoluene, 2,6-dinitrotoluene, 1,3,5,7-tetranitrooctahydro-1,3,5,7-tetrazocine (HMX), nitrobenzene, and tetryl.

[3]No data were available between 1990 and 1995.

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[3]No data were available between 1990 and 1995.

collected for each vertical profile. Up to 400 g of soils were collected in the sampler.

Explosives concentrations determined with a field test kit (Ensys, Inc., Research Triangle Park, NC) (Jenkins, 1990; Walsh and Jenkins, 1991) were validated with results obtained in the laboratory. Explosives and geochemical parameters, including permeability, were measured on these samples that were also used for biomarker research. Soil samples were extracted and analyzed for explosives and transformation products of explosives by Method 8330 (USEPA, 1988). Soil samples were also analyzed for pH (Mehlich, 1984), nitrate nitrogen, nitrite nitrogen, total Kjeldahl nitrogen, phosphorus, sulfate, and total organic carbon (American Public Health Association, 1985). Hydraulic conductivity was determined by standard sieve methods. Particle size distribution in the < 2 mm fraction was determined by the methods of Day (1956) as modified by Patrick (1958).

The CPT data defined the Area P deposits as predominately silts, clays, and silty sands. Explosives detected were primarily limited to TNT, RDX, TNB, and HMX with sporadic detections of DNB, 4-amino-2,6-dinitrotoluene (4ADNT), 2-amino-4,6-dinitrotoluene (2ADNT), 2,4DNT, and hexahydro-1-nitroso-3,5-dinitro-1,3,5-triazine (MNX). The highest concentration detected was 13.1 mg TNB kg^{-1} at one location. Maximum concentrations for TNT and RDX were 7.1 and 11.8 mg kg^{-1}, respectively. The DNB and 2ADNT were restricted to two locations. The detection of 4ADNT was limited to one location. The 2,4DNT was detected in only two transects, at two locations on each. The MNX was observed at three depths in one location. No diamino transformation products of TNT were observed. Generally, explosives concentrations increased with depth until reaching a maximum, and then decreased, indicating passage through a single plume of contamination (Figure 5). No significant relationship (as determined by Pearson Product Moment Correlation) could be established between concentrations of TNT, RDX, and TNB and cone penetrometry tip resistance (an indicator of the kind of material penetrated) (Figure 5).

Groundwater Monitoring

Well selection criteria included appropriate spatial distribution of wells relative to the original source and relative to the existing conceptualization of the contaminant plume (SAIC, 1994), proximity to potential receptors or site boundaries, and availability of well completion data concerning screen type, depth, and well performance, such as water yields. Thirty wells were selected (Figure 1). During the first 6 months, monitoring was scheduled monthly in order to refine the sampling techniques and sample handling procedures. After the first 6 months, sampling was reduced to quarterly for the duration of the 2-year demonstration period. After the first year, the number of wells was limited to 16 to reduce the frequency of repeated monitoring of wells that had been consistently uncontami-

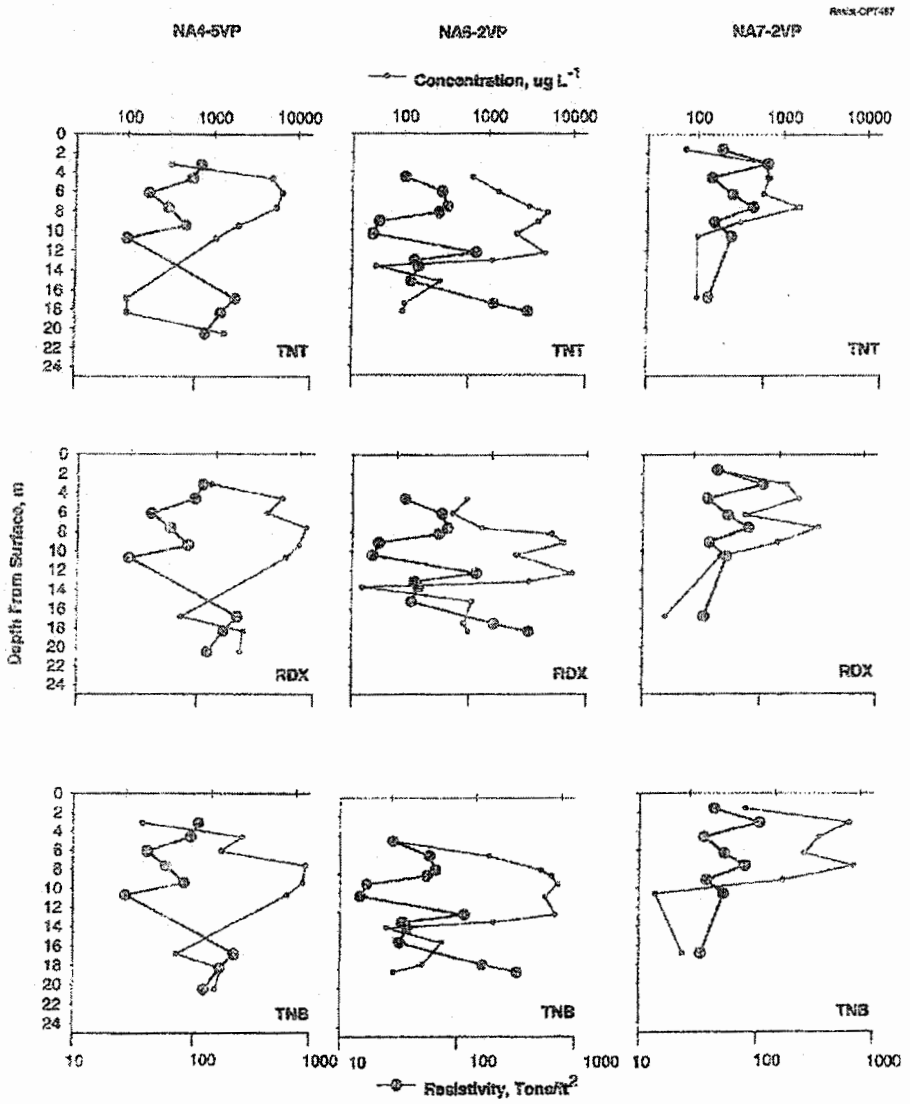

FIGURE 5

Concentrations of explosives (TNT, RDX, and TNB) and cone penetrometry tip resistivities with depth at three LAAP locations.

nated and were not within the immediate vicinity of the contaminant plume. All 30 wells were sampled again at the end of the second year.

Groundwater Sampling Methods

Wells were sampled by micropurge (low-flow) techniques (Gass *et al.*, 1991). Field parameters were measured with an in-line continuously monitoring unit (Yellow Springs Instruments, Yellow Springs, MO) with data transmitted directly to a laptop computer. Parameters were recorded in real time. These parameters included pH, conductivity, dissolved oxygen (DO), temperature, redox, turbidity, and salinity. Samples were taken after a stable DO reading was obtained while matching discharge to recharge. Dissolved oxygen stability was defined as achievement of two successive readings within 10% of each other. When recharge was too slow to use the micropurge technique, the well was evacuated for 10 min and allowed to recharge three consecutive times before sampling.

Groundwater was directed through a 0.45-μ filter into separate bottles already containing appropriate preservatives for the specific analyses. During month 3, a sample from each well was assayed for picric acid. Iron speciation, Fe^{+2} and Fe^{+3}, and methane were assayed in all wells in month 10 and in 12 selected wells in month 12. Samples for speciation were collected by bailing after samples for explosives and other geochemical parameters had been collected by pumping. Samples were transferred from the bottom of the bailer into volatile organic analysis tubes to reduce contact between the sample and air. All samples were stored on ice or under refrigeration until analyzed. All monitoring wells, physical boundaries and features, and CPT and surface soil sampling locations were surveyed using a global positioning system.

Analytes and Analytical Methods

Due to the several potential attenuation processes affecting explosives, analysis of a battery of parent compounds and transformation/degradation products was necessary (Table 2). Some geochemical parameters were monitored in the field, while others were assayed in the laboratory (Table 3). Attenuation processes are typically too slow for accumulation of sufficient degradation products (e.g., nitrates, ammonia, methane, and carbon dioxide) to be distinguished from background. However, the geochemical properties contribute to the weight-of-evidence that conditions in the site are suitable for attenuation processes.

Data Quality Assurance

An important objective of the project was to optimize groundwater data quality by minimizing random error and systematic bias. Therefore, a comprehensive quality

Table 2

Explosives and Related Analytes Monitored in Groundwater at LAAP and JAAP

Chemical Name	Acronym
2,4,6-Trinitrotoluene	TNT
1,3,5-Trinitro-1,3,5-hexahydrotriazine	RDX
Octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine	HMX
N,2,4,6-Tetranitro-N-methylaniline	Tetryl
1,3,5-Trinitrobenzene	TNB
2,6-Dinitrotoluene	2,6DNT
2,4-Dinitrotoluene	2,4DNT
1,3-Dinitrobenzene	1,3DNB
Nitrobenzene	NB
4-Amino-2,6-dinitrotoluene	4ADNT
2-Amino-4,6-dinitrotoluene	2ADNT
3,5-Dinitroanaline	35DNA
2,4-Diamino-6-nitrotoluene	24DANT
2,6-Diamino-4-nitrotoluene	26DANT
2,2',4,4'-Tetranitro-6,6'-azoxytoluene	66'AZOXY
4,4',6,6'-Tetranitro-2,2'-azoxytoluene	22'AZOXY
2,2',6,6'-Tetranitro-4,4'-azoxytoluene	44'AZOXY
Hexahydro-1-nitroso-3,5-dinitro-1,3,5-triazine	MNX
Hexahydro-1,3-dinitroso-5-dinitro-1,3,5-triazine	DNX
Hexahydro-1,3,5-trinitroso-1,3,5-triazine	TNX
Mononitroso-octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine	MN-HMX
Picric acid	

Table 3
Geochemical Parameters Monitored in Groundwater at LAAP and JAAP

Monitored in the Field	Assayed in the Laboratory	
	Aerobic Collection	Anaerobic Collection
pH	Total organic carbon	Reduced iron
Conductivity	Total iron	Methane
Dissolved oxygen	Calcium	
Temperature	Magnesium	
Redox potential	Manganese	
Turbidity	Nitrate/nitrite nitrogen	
Salinity	Sulfate	
	Chloride	

assurance plan was constructed, including some initial experiments, to optimize the following: standard operational procedures for sample collection and preservation, precision, accuracy and representativeness of the data, data comparability, and field quality control.

Between well sampling events, the groundwater in the immediate vicinity of the well head has the opportunity to become oxidized due to contact with the air column in the well. The extent of the oxygenated zone around the well head is a function of such factors as local permeability of the aquifer, the flow rate of the groundwater, and the time since last well purging. Dissolved oxygen concentrations in this zone may promote higher microbial activity than is typical in the groundwater beyond the well. Such activity may significantly affect the contaminant by promoting degradation or transformation. Because the oxygenated zone may not be representative of the aquifer, artifacts may be introduced into the data if this water is not removed before sampling. In order to test the relationship between dissolved oxygen and contaminant concentrations, samples from three wells were collected every 10 min until the dissolved oxygen readings were stable. TNT concentration was inversely correlated with dissolved oxygen concentrations over pumping time in two of the wells ($r^2 = -0.76$ and -0.94; $p = 0.019$ and 0.001 for each well, respectively). In the third well, the relationship was inverse ($r^2 = -0.49$), but not significant ($p = 0.220$). The RDX concentration was consistently inversely correlated with dissolved oxygen, but significant in only one well ($r^2 = -0.95$, $p < 0.001$). Several transformation products of TNT also exhibited significant negative correlations with dissolved oxygen.

A second experiment assessed whether sample preservation was essential to prevent analyte loss during the period between sample collection and analysis. Samples were collected from 10 wells and each was split into three aliquots; a control, which was not chemically preserved, an aliquot acidified to pH 2 with 1.5 g sodium bisulfate ($NaHSO_4$) L^{-1}, and one preserved by addition of 60 mg mercuric chloride ($HgCl_2$) L^{-1}. Samples were stored on ice and analyzed for explosives. Results of a two-way analysis of variance (ANOVA) with preservative as the treatment variable and monitoring well as a blocking variable indicated no differences among preservatives even after accounting for concentration differences between wells.

One of every 10 samples was collected and analyzed in duplicate to estimate precision of the data, including contributions from sample collection, storage, preconcentration, and determination. No differences between analyte concentrations for duplicate samples in a paired t-test indicated that precision was good. Accuracy was estimated from spiked samples subjected to the entire analytical process. Two spiked blanks and three spiked groundwater samples, both low and high concentrations, were analyzed for each quarterly sampling event. The accuracy of the analytical methods was typically within two standard error units of 100%. No analytes were detected in unspiked blanks.

One of every 10 samples was split and analyzed independently by the U.S. Army Cold Regions Research and Engineering Laboratory (CRREL) using the same analytical method used by the WES Environmental Chemistry Branch. Results of a paired t-test indicated no difference between analytical results achieved by the two laboratories for six of the seven analytes. CRREL values for HMX tended to be greater than WES values ($P = 0.002$).

Because concentrations of explosives ranged from less than the analytical detection limits (0.2 $\mu g\ L^{-1}$) to more than 20 $mg\ L^{-1}$, or about five orders of magnitude, the potential for carryover of explosives as sampling progressed from well to well from high to low concentration was great. Therefore, wells were sampled in order from lowest to highest concentrations. To further minimize the opportunity for carryover, Teflon tubing was dedicated to each well. The sampling pump was purged in clean water between wells. One sample of rinsate from equipment decontamination was analyzed for explosives each sampling day.

Data comparability was achieved by following the sampling protocol consistently for each well in each sampling event and implementing the quality assurance plan described above. Data comparability was stressed during field sampling and chemical analyses to optimize detection of differences in explosives concentrations as a function of time.

Statistical Analyses

Trend analysis was performed on the 2-year groundwater contaminant data using the SAS LIFEREG procedure (SAS Institute Inc., 1988) assuming either a normal or a lognormal distribution. The LIFEREG procedure fits a regression line by maximum likelihood estimation (MLE) and is particularly well suited to analysis of censored data such as those with below detection limit observations. LIFEREG incorporates probabilities below detection limit for the assumed distribution and does not require prior reconstitution of the censored data (e.g., using one half the detection limit). However, a simulation study in progress has shown the LIFEREG procedure to have a high Type I (false-positive) error rate in some circumstances. Therefore, trend analysis was also performed by ordinary least squares (OLS) regression using linear, logarithmic, and quadratic models in the SAS REG procedure, following substitution

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of one half detection limit for less than detection limit observations. Trends were not considered valid unless significant ($p < 0.05$) by at least one of the MLE models and one of the OLS models. As a check on the validity of the regression trend analyses, nonparametric Spearman's rank correlations were performed, correlating contaminant concentrations with sample time in each well. Trends in contaminant concentrations over the 2-year study period were analyzed statistically for the 11 wells in which most analytes were consistently detected. Significant declines in contaminant concentrations occurred in 9 of the 11 wells. (Figure 6 illustrates trends in explosives concentrations over time in four of the most contaminated wells.)

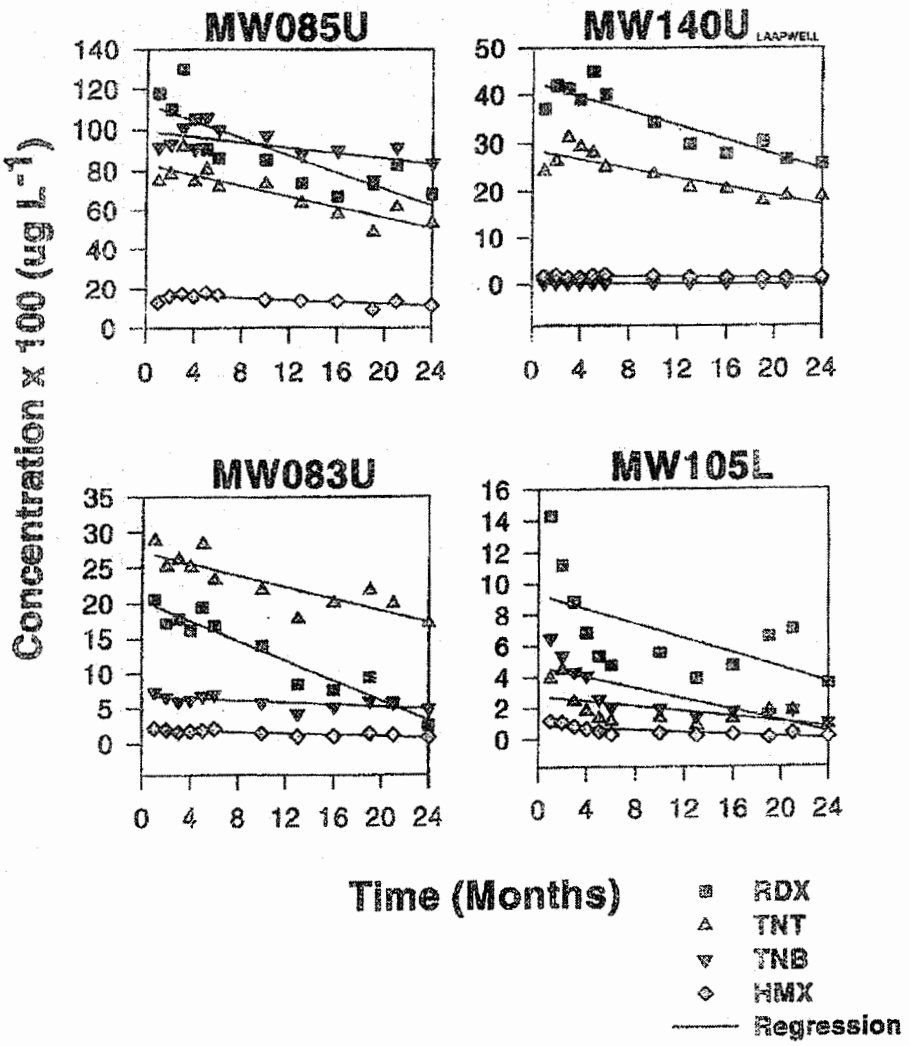

FIGURE 6

Trends in explosives concentrations over sampling events at LAAP.

Contaminant concentration data were correlated with concurrent geochemical parameters (Ca, Cl, Fe, Mg, Mn, NO_2/NO_3, SO_4, TOC, and water level) when the data for a given well included at least three detectable concentrations of both the contaminant and the geochemical parameter. Both Pearson's *r* and Spearman's nonparametric correlation analyses were conducted because so many explosives values were less than the detection limit and because sample size was relatively small ($n = 5$ to 12). Correlations were considered significant only when both analyses produced significant results ($p < 0.05$). Few significant correlations between explosives concentrations and Ca, Cl, Fe, NO_2/NO_3, SO_4, and water level were observed. Several significant correlations were observed between explosives concentrations and Mg and Mn, and these were predominantly positive. The TOC was positively correlated with most of the explosives concentrations in most of the wells. These results suggest that geochemical characteristics are generally not consistently related to explosives concentrations.

Estimates of Site Capacity for Attenuation

Capacity for adsorption and transformation can vary considerably across a site; therefore, accurate lithological profiles and determination of sorption and transformation parameters on representative aquifer soil samples is essential. Batch tests with LAAP soils followed standard procedures (40CFR796.2750 http://frwebgate.access.gpo.gov/cgi-bin/multidb.cgi at 40CFR796.2750-- Sec. 796.2750 Sediment and soil adsorption isotherm). The four soil types identified in the LAAP aquifer were generally high in sand, ranging from 65 to 92.5% sand. Silt and clay was present in all samples, although in smaller quantities than sand. Total organic carbon content was low, ranging from 0.015 to 0.162%. The cation exchange capacity (CEC) was also low, ranging from 3.5 to 8.1 Meq 100 g^{-1}. Soil pH was acidic and relatively consistent for all soil types (average of 5.55). Permeabilities of the soils ranged from 10^{-4} to 10^{-9} cm sec^{-1}. Most explosives reached steady state distribution between aquifer soils and water within 24 h (Pennington *et al.*, 1999), making equilibrium partitioning an appropriate assumption for sorption of most of the explosives at this site.

Adsorption of explosives from groundwater by the LAAP aquifer soils was limited. The measured values of K_d were below 1 L kg^{-1} for all soils and contaminants, ranging from no significant adsorption to a high value of 0.84 L kg^{-1}. The highest degree of sorption was associated with the soils highest in clay and CEC. The range of sorption coefficients between soils varied over an order of magnitude for explosive compounds. For modeling of contaminant transport at the LAAP site, the use of an average value of K_d to represent sorption was appropriate. Although these data indicated limited retardation, low permeability may exert a greater influence on movement of the plume than is exerted by sorption and transforma-

tion. These results suggest that mass transport limitations rather than site capacity restrict transport at LAAP.

Biomarker Analyses

Three sets of tests were evaluated to determine their usefulness in supporting site evaluation of monitored natural attenuation. These included (1) soil mineralization radioassays, (2) lipid biomarker analyses, and (3) nucleic acid biomarker analyses. These tests were designed to evaluate the microbial degradation potential of the site. For the radioassays soils from the aquifer received radiolabeled acetate, TNT, or RDX in slurry reactors. Evolution of radiolabeled CO_2 from acetate indicated the general activity of the site microflora, while evolution of radiolabeled CO_2 from the explosives indicated degradation potential and provided an estimate of rate. Lipid analyses provide quantitative data on the *in situ* viable microbial biomass, community composition, and physiological status. For the lipid assays ester-linked phospholipid fatty acids (PLFA) were extracted from aquifer soil, fractionated on a silica gel column, and analyzed by gas chromatography. PLFA data were quantified by comparison with known internal and external standards and from patterns and types of PLFA profiles determined from a growing database. For nucleic acid assays DNA was isolated from the aquifer soils, genes encoding for enzymes involved in degradation of nitroaromatic compounds were amplified, and diagnostic banding patterns were determined by slab gel electrophoresis. The presence of the targeted catabolic genes was correlated with the radioassay results to establish a link between the presence of the requisite genes in the soil and the observed mineralization activity.

The half-life of TNT by mineralization ranged from 320 to more than 3,900 days; for RDX the range was from 323 to more than 3900 days. Few significant correlations between geochemical parameters and biomarkers were found. Several nucleic acid probes correlated positively with mineralization rate, as did the following parameters determined by lipid biomarkers: biomass, abundance of Gram-negative, sulfate-reducing, and iron-reducing bacteria, and a sulfite reductase. The potential for aerobic degradation of TNT in LAAP soils was suggested by the presence of two catechol oxygenase gene probes. Other observed genes supported potential for both anaerobic and aerobic metabolism of TNT.

Stable Isotope Analyses

The objective of this part of the study was to demonstrate the feasibility of using stable isotopic ratios of carbon ($\delta^{13}C$) and of nitrogen ($\delta^{15}N$) in either soil or groundwater to monitor natural attenuation of explosives. Two strategies were explored, changes in carbon isotopic ratios in the soil, and changes in nitrogen

isotopic ratios in groundwater. Measuring changes in carbon isotopic ratios in groundwater is not practical because of the limited dissolved organic carbon typically present. However, the incorporation of TNT or covalent bonding of amino-daughter products of TNT to soil may result in a change in the $\delta^{13}C$ of the soil. Therefore, the detection of such changes in soil may be feasible. Measuring changes in $\delta^{15}N$ in soil is not practical because of the many confounding sources of nitrogen present. However, measuring changes in $\delta^{15}N$ of TNT from groundwater is promising. The bonding of the amino reduction products of TNT to the soil humic material is through the amino functional group (Thorn, 1998). The reduction of nitro groups, whether chemical or biological, may result in isotopic fractionation due to reaction-rate differences between ^{15}N and ^{14}N. If so, a relatively greater percentage of ^{14}N should be reduced and involved in reactions with either humic material or clay. This process would leave relatively more ^{15}N in the TNT remaining in the groundwater.

Two laboratory and two field studies were conducted. In the first laboratory experiment, the $\delta^{13}C$ and $\delta^{15}N$ values for TNT were measured over time when a known amount of TNT was added to wet soil in a test tube under static conditions. In the second laboratory experiment the $\delta^{13}C$ and $\delta^{15}N$ values for TNT were measured in a soil/water system where an unlimited amount of TNT was made available to the system by periodically replacing the solution phase with fresh TNT-containing water. One of the two field studies analyzed stable isotope ratios of carbon in the extractable TNT from soil core samples collected at different depths and across the plume by cone penetrometry. In the second field study groundwater samples collected along transects across the contaminant plume were analyzed for TNT concentration and for the $\delta^{15}N$ and $\delta^{13}C$ values in the extractable TNT.

Results of the first laboratory experiment demonstrated that although the concentration of extractable TNT decreased during incubation, no changes could be detected in the $\delta^{13}C$ value for the soil. The total percent carbon in the soils in this experiment ranged from 0.012 to 1.3%. If TNT became bound to the soil organic matter at a level of 0.2% (w/w), the mass of carbon resulting from the TNT would only be 19 μg in a 2-g soil sample. This represents only 0.074% of the total carbon present in the sample, which generates an isotopic ratio difference too small to measure. Results of the second laboratory experiment demonstrated a definite and dramatic decline in TNT concentration over time; however, changes in both isotopic ratios were inconclusive due to the limited precision of the measurements.

TNT was detected in most soil cores at concentrations ranging from 0.1 to 1.8 μg/g. Analysis indicated no difference between the $\delta^{13}C$ values for uncontaminated and TNT-contaminated soils. The extractable TNT was also analyzed for $\delta^{15}N$; however, due to the low concentration of TNT in the extracts, reliable $\delta^{15}N$ measurements could not be obtained. No significant differences in the $\delta^{13}C$ values for TNT extracted from groundwater at LAAP were observed. However, the $\delta^{15}N$ values of TNT in groundwater samples increased from the center of the plume

outward. A Student's t-test showed a clear trend of increasing $\delta^{15}N$ values for TNT across the plume. Therefore, TNT is isotopically fractionated as it moves through the subsurface. These results suggest attenuation of the lighter isotopic fraction of TNT leaving the groundwater enriched in the heavier isotope. Therefore, using $\delta^{15}N$ measurements of TNT from groundwater to monitor natural attenuation of TNT is promising.

Groundwater Modeling

The Department of Defense Groundwater Modeling System (GMS) version 2.0 (GMS, 1996) was applied to the LAAP site using information derived from site monitoring, site capacity estimates, and biomarker analyses. The objectives of the numerical modeling effort were (1) to visualize contaminant distribution at the site, and (2) to predict long-term contaminant migration and/or attenuation. The modeling consisted of conceptual model development, calibration of input data, sensitivity analysis, and long-term predictions (Pennington *et al.*, 1999).

The modeling domain at the ground surface of the LAAP site included the former lagoon area and proximal monitoring wells. Four stratigraphic units were identified at LAAP, a vadose zone (unsaturated soil), an upper terrace aquifer, a semiconfining layer, and a lower terrace aquifer. Values of hydraulic conductivity for each layer and node of the three-dimensional modeling mesh were estimated from discrete hydraulic conductivity data (cone penetrometry) using GMS geostatistical tools. Water level elevations, which were measured at the monitoring wells, were used to estimate transient flow boundary conditions. The source of flow recharge at Area P was assumed to be solely from rainfall. Therefore, rainfall data were used to estimate the average infiltration rate for the site.

The model application is limited by the assumptions applied to the numerical model, which include laminar flow conditions, incompressible fluid and solid phases, and constant fluid viscosity and density. The input data for FEMWATER include hydraulic conductivity, porosity, hydraulic gradient, infiltration rate, initial and boundary conditions, distribution (partition) coefficient, and decay rates. Results from laboratory batch testing for adsorption of explosives (TNT and RDX) to LAAP soils were used to develop retardation factors. An average value of 0.23 L kg^{-1} for TNT and 0.30 L kg^{-1} for RDX was derived from the empirical determinations on four samples of LAAP aquifer soils. Decay rates were based on the radiorespirometry results and model calibration using contaminant concentrations measured in the field. The values were 10^{-5} per day (half-life of 190 years) for TNT and 8.13×10^{-6} per day (half-life of 233 years) for RDX.

Sensitivity to changes in the values of hydraulic conductivity, adsorption coefficient, and decay rate was evaluated. Increasing or decreasing the hydraulic conductivity by 10% did not significantly impact the modeled results. The apparent lack of sensitivity to hydraulic conductivity is likely due to the relatively low

conductivity associated with this site. One order of magnitude change in the adsorption coefficient caused a 40% change in TNT concentration at the end of 1000 days of the simulations. This indicated that the model is sensitive to the adsorption coefficient. The extent of adsorption on aquifer soils was quite low relative to surface soils (Pennington and Patrick, 1990). The decay rate was varied from 10^{-5} to 10^{-4} day^{-1}, while holding adsorption and conductivity constant. A 12% change in the predicted TNT concentration at the end of 1000 days simulations indicated that the model is moderately sensitive to the decay rate.

To predict changes that might occur in the distribution of TNT and RDX after 20 years, the following assumptions were imposed on the model: (1) no additional source of contamination is added into the site, (2) infiltration rate is constant throughout the time period under investigation, (3) flow boundary conditions recur every year, and (4) no recharge or discharge through pumping occurs during the simulations.

When initial conditions (Figures 2, TNT) were compared with simulations for 20 years (Figure 7, TNT), the areal extent of both plumes was diminishing. The highest concentration was reduced from 10,500 to 250 $\mu g\ L^{-1}$ for TNT and 23,200 to 620 $\mu g\ L^{-1}$ for RDX. Contaminant masses were calculated using the measured and predicted TNT and RDX concentrations along with iso-surface volume calculation of GMS. The estimated initial masses of TNT and RDX (based on the first groundwater monitoring event of the project) were 52 and 78 metric tons, respectively. The predicted values after 20 years were 7 and 4 metric tons for TNT and RDX, respectively. The accuracy of these numbers is controlled by the assumptions and limitations of FEMWATER and by the iso-surface calculations of GMS.

Conclusions

Results of the groundwater monitoring over 2 years at LAAP demonstrated declines in explosives concentration at specific points in the plume and reduction in total estimated contaminant mass. None of the geochemical parameters correlated with declines in explosives concentrations. Therefore, most of these parameters probably need not be monitored. Initial site characterization should include particle size distribution, organic carbon, NO_2/NO_3, and Fe, parameters that may contribute to a weight of evidence for attenuation by establishing characteristics of the site that make it suitable for attenuation processes.

Site capacity data showed that mass transport limitations rather than site capacity for attenuation restrict transport at LAAP. Estimation of site capacity should be determined for each site due to the differences in site-specific hydrogeology and geochemistry. Results of biomarker assays demonstrated the potential for microbial degradation processes at the site and contributed to a weight-of-evidence for these processes. The only microbial technique sufficiently developed to recommend is radiorespirometry with aquifer soil from the site. Lipid and nucleic acid

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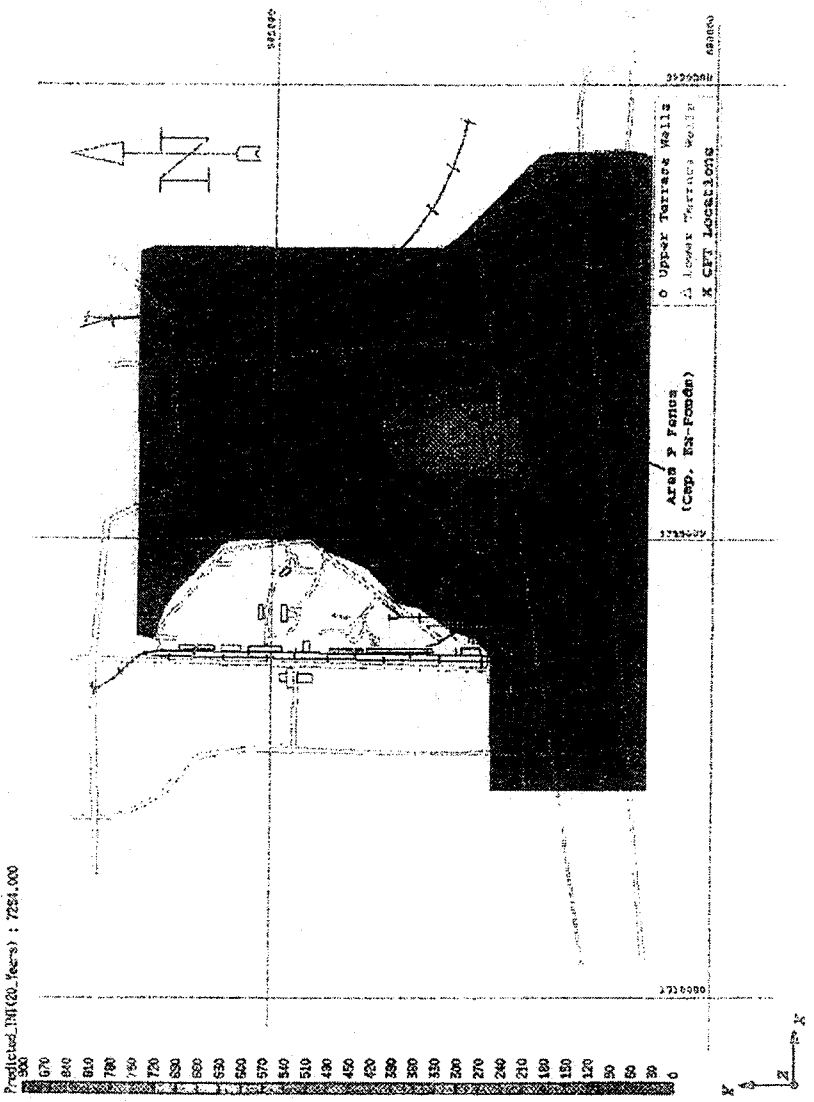

FIGURE 7

GMS prediction of extent of TNT groundwater plume after simulation of 20 years. Units on vertical scale are ppb.

biomarkers showed promise, but require further development before they can be recommended for general use. Results of stable isotope assays suggest that $\delta^{15}N$ in groundwater may be useful for tracking the attenuation process, but this approach also requires further development.

The numerical model predicted continuing declines in explosives mass over time. This information can be used to predict when site clean-up goals may be achieved. Even though attenuation processes are slow, the site hydrogeology at LAAP provides sufficient time for attenuation processes to protect potential receptors. Each site will require careful development of a mass of data on which a decision to use monitored natural attenuation can be based.

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