



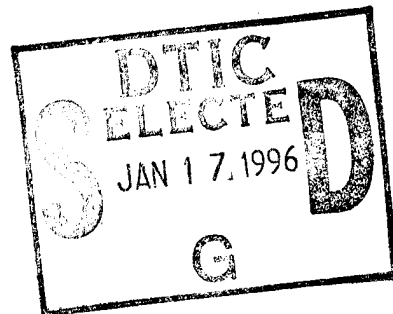
# EDGEWOOD

RESEARCH, DEVELOPMENT & ENGINEERING CENTER

U.S. ARMY CHEMICAL AND BIOLOGICAL DEFENSE COMMAND

ERDEC-TR-268

## TOXICITY OF SEDIMENTS SURROUNDING THE GUNPOWDER NECK SUPERFUND SITE AT APG, MD



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July 1995

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13. ABSTRACT (Maximum 200 words) From the late 1940s through the 1960s, the standard practice for disposing of toxic chemicals at Aberdeen Proving Ground, MD, was open burning. Historic records show that an open burning site was used to dispose of a variety of Army unique chemicals. This disposal site has since been placed on the National Priority List (NPL) by U.S. Environmental Protection Agency. In the early 1980s, several environmental surveys were conducted that included soil sampling for contaminants and installation of wells for monitoring ground water contaminants. In the spring 1992, sediment samples were taken from waterways that surround that disposal area. Chemical analysis and sediment toxicity assays ( <i>Ampelisca abdita</i> ) were conducted. Toxicity comparison, with sediment leachate from an Adapted Toxicity Characteristic Leaching Procedure (ATCLP), were made using <i>Daphnia magna</i> and a fluorescent bacterium <i>Photobacterium phosphoreum</i> in MICROTOX assays. Amphipods showed a wide range of mortality in mud as well as coarser sediments indicating substrate preference is not critical to the outcome of the assay. Toxicity results from the leachates showed the sediments were not toxic to daphnia and MICROTOX assays.				
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PREFACE

The work described in this report was authorized under Project No. 10162622A553, CB Defense/General Investigation. This work was started in August 1992 and completed in December 1993.

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## QUALITY ASSURANCE

This study, conducted as described by Protocol 22092000X047, was examined for compliance with Good Laboratory Practices as published by the U. S. Environmental Protection Agency in 40 CFR Part 792 (effective 17 Aug 1989). The dates of all inspections and the dates the results of those inspections were reported to the Study Director and management were as follows:

<u>Phase inspected</u>	<u>Date</u>	<u>Date reported</u>
Sediment prep, amphipod addition	20 Aug 92	24 Aug 92
Amphipod retrieval, counting	31 Aug 92	2 Sep 92
Data & Final Report	29 Jun 94	29 Jun 94

To the best of my knowledge, the methods described were the methods followed during the study. The report was determined to be an accurate reflection of the raw data obtained.

  
DENNIS W. JOHNSON 21 Jun 94  
Quality Assurance Coordinator  
Research and Technology Directorate

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TOXICITY OF SEDIMENTS SURROUNDING  
THE GUNPOWDER NECK SUPERFUND SITE AT APG, MD

1. INTRODUCTION

The J-field disposal site, located at Aberdeen Proving Ground (APG), MD, was used from the late 1940s through the 1960s to dispose of a variety of Army unique chemicals, laboratory waste, pilot plant operations, and high explosive rounds through open burning or detonation.<sup>1,2</sup> Table 1 lists the documented chemicals that were disposed in the J-field area before its closure.<sup>3,4</sup> The J-field site (pits and surrounding area) consists of 135 acres of woodland, meadows, and marshland at the end of Gunpowder Neck surrounded by the Bush and Gunpowder Rivers (Figure 1). The disposal area, on the tip of the peninsula, consisted of pits ranging in size from 300 ft by 15 ft to the smallest being only 4 ft by 6 ft. Typically, open trenches or pits were excavated, filled with wood (kindling), and soaked with fuel oil. Containers of material were placed on top of the wood and the fuel ignited. The containers were breached using an explosive charge, allowing the disposal materials to be incinerated in place. The Environmental Protection Agency (EPA) has placed this site on the National Priority List (NPL).

From the late 1970s into the 1980s, several environmental surveys were conducted, which included soil sampling for contaminants and installation of wells for monitoring ground water contaminants.<sup>1</sup> Terrestrial soil samples collected from the toxic pit areas in 1986 contained concentrations of arsenic, barium, chromium, lead, mercury, silver, and cadmium. Unfortunately, no background analysis was available to determine concentrations of the metals before disposal operations started. The soil samples also contained concentrations of polychlorinated bi-phenols, naphthalenes, hydrocarbons, chlordane, and various pesticides (DDT, DDD, and DDE). Ground water samples were analyzed and low concentrations of organics such as chlorobenzene, ethanes, ethylenes, toluene, and chloroform were detected.

The past environmental surveys have been concerned with the contamination at J-field. The major concern now is whether contaminants at the site could be leaching into off-shore sediments, potentially impacting the biota and creating potential hazards to commercial and recreational activities around the vicinity of J-field.

The purpose of this study was to conduct toxicity assays with off-shore bottom sediments from the tributaries and bay that surround J-field. The marine amphipod *Ampelisca abdita* was used in sediment toxicity tests, coupled with chemical analysis, to gain insight as to the toxic effects the bottom sediment may be having on the ecosystem. *Daphnia* and MICROTOX assays were also conducted using extracts from the sediment samples to cross reference toxicity results.

## 2. METHODS AND MATERIALS

The flow chart in Figure 2 briefly describes the testing scheme used in assessing the toxicity of sediments taken from tributaries surrounding the J-field disposal site.

Roy F. Weston, Incorporated, (Edison, NJ) was contracted to provide APG with a total of 16 samples taken from around the Gunpowder Neck area of the Gunpowder and Bush Rivers (see Figure 1). One sample (#17) from the Fairlee Creek, located on the eastern shore of the Chesapeake Bay, directly across from the J-field area, was also taken to serve as a reference site. The sampling operation included scanning the river bottom with magnetometers seeking unexploded munitions, collecting the sediments using a remote sampling barge and controlling the chain of custody in the distribution of sediment samples.\*

After the sediments were collected, they were screened for mustard and nerve agent contamination through head space analysis techniques, (completed by the Chemical Support Division, Chemical and Biological Defence Agency, APG). Once cleared, the sediment was transported to various destinations for further analytical and environmental studies. Roy F. Weston, Incorporated, provided additional chemical analyses (explosives, agent degradation products, metals, PCBs, pesticides, and chemical surety materials) and grain size analysis.\*

### 2.1 In-House Analytical Determinations.

Because the detailed chemical analysis was conducted the same time as the sediment toxicity assays, a Bruker Mobile Mass Spectrometer (MM-1) was used to assess the sediments for volatile organic materials that may constitute worker health hazard.

The MM-1 was used to perform numerous environmental sampling with an enhanced extended data station that provided significant unknown search and library match capability. This instrument package is mounted inside a cargo van and is fully operational in the field using an on-board power supply. The environmental probe head include a heated surface probe, a thermal desorption gas chromatographic oven, and a water probe for in place analysis of organics from water matrices.

Internal function, electronic, temperature, and calibration tests were conducted before sample analysis. Automatic calibration allows the instrument to calibrate on approximately ten different masses over the instrument's detection range (45-400 amu). Because filtered air is used as a carrier gas, sensitivity of the instrument is checked by focusing on the profile spectrum of a substance already present in the air (argon, mass units = 40). Log scale readings of 6.5 for argon represent very good sensitivity readings. However, the sensitivity reading was nearly 7.0 during

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\*Noel T. Rogers, Roy F. Weston, Incorporated, GSA Raritain, Depot, Edison, NJ, provided analytical work for this study.

the sediment sample analysis. Another check is performed by observing instrument response to a standard benzene/toluene/xylene mix, which is compared to manufacturers specified responses.

The sediment samples were analyzed using the heated surface probe. The probe is a flexible 3 m sampling device containing a 10 m column that directly attaches to the entrance of the mass spectrometer. During initial analysis, the probe head was maintained at 240 °C and the column at 200 °C. Because the spectra were collected every second, increases above baseline could be quickly detected. The samples were then re-analyzed by subjecting the sample to a temperature program and obtaining more accurate chromatographic data on the samples of interest.

Sediment samples were placed on a flat glass surface. The probe head was placed on the sample for 15-20 s to allow any volatile or semi-volatile components to vaporize. After collection of the data by the external data system, the sample head and glass surface were cleaned with deionized water to remove any residual materials.

## 2.2 Sediment Toxicity Assay.

Marine amphipods (*Ampelisca abdita*) were collected from sites located in Narraganset, RI.\* The organisms, native sea water, and sediment (control) were shipped via overnight freight. The organisms were placed in 10-gal glass aquaria with 4 cm of sediment and allowed to acclimate to test conditions for 48 hr.

Amphipods were removed from the acclimation aquaria by collecting the bottom sediments and sifting the mud and debris through a 0.5 mm stainless steel sieve. The animals were placed in holding chambers containing fresh sea water. Immature amphipods of uniform size were used in testing. The animals were transferred sequentially, via pipette, to sample cups (20 animals each) to reduce bias in animal choice, and then each cup was then randomly assigned to a test chamber.

The test chambers consisted of 1 L pre-washed EP glass jars (See Appendix A for the washing procedures). The test chambers and lids were rinsed with sea water before adding the sediments. As a safety precaution, all sediment transfers were conducted under a certified chemical exhaust hood. Control sediments from Narraganset were passed through a 2 mm stainless steel sieve to eliminate debris and any large predators. However, the sediments from the J-field area consisted of sand, clay, and detrital mud, which would not pass through the sieve (the Fairlee Creek control sediment also could not be sieved). Sediment was transferred into the test chambers to provide approximately 4 cm of depth (200 mL). During sediment manipulations, the MM-1 was used to insure the sediments were clear of possible harmful volatile materials not screened for during the initial agent screening process. Dilution water was added to the test chamber (approximately 800 mL), trying

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\*Science Application International Corporation, Narraganset, RI, provided test organism for studies.

not to disturb the sediment. The test chambers were covered and gently aerated (not to cause the sediment to re suspend) with a pipette inserted through the lid, to approximately 1-2 cm above the sediment surface for 24 hr. After the aeration period, the test chambers were randomly placed in a temperature controlled room (20 °C), with a light dark cycle of 16/8 hr.

When the amphipods were to be added to the test chambers, aeration was stopped and the contents of the sample cups containing the amphipods were poured through a 0.5 mm stainless steel screen to reduce the water additions, recounted, and rinsed off the screen into the test chambers. Any floating amphipods were gently pushed under the water surface with a pipette. The animals were allowed 1 hr to burrow in to the mud. Any amphipods not burrowed were removed and replaced with another and aeration resumed.

Daily observations were conducted looking for floating or emerged animals. Floating animals (live) were noted and submerged. Any motionless animals seen on the water or sediment surfaces were removed and examined under a dissecting microscope to determine mortality. If any signs of life (neuromuscular twitching) was observed, they were returned to the test chamber and gently submerged into the sediment.

At the end of 10 days, the test was terminated and mortality was determined. The contents of the test chambers were rinsed individually through 0.5 mm sieve, using sea water to remove mud and expose the amphipods. If 16 out of 20 animals were not found, including any dead animals reported during the test, the sample was examined by another technician for quality control purposes. All missing animals were considered dead.

Due to the number of replicate chambers, several of the test chambers had to be preserved and mortality determinations made at a later date. To avoid bias mortality determinations, only two replicates from each sediment sample were preserved. Therefore, live mortality determinations were made on three out of five replicates from each sediment sample. The preserving process consisted of sieving the contents of the test chambers as described above. The remaining debris was placed back into the test chamber, and the chamber filled with enough 10% formalin live-stain (8.0 g mono-basic  $\text{NaPO}_4$ , 12.6 g dibasic  $\text{Na}_2\text{PO}_4$ , 1800 mL distilled water, and 200 mL formaldehyde, Rose-Bengal until the solution became a deep cranberry red)\* to cover the debris remaining in the jars. Live amphipods stain bright pink when preserved, thus identifying viable organisms present at the end of the test. Before the mortality determination could be made on the preserved samples, they were rinsed into a waste containment system several times in a chemical hood to avoid worker exposure to formalin. Mortality determinations were then recorded as described above.

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\*M.S. Redmond, K.J. Scott, K.M. McKenna, and D.L. Robinson, Science Application International Corporation, Narragansett, RI, provided Test Methodology Reference for this study.

### 2.3 Adapted Toxicity Characteristic Leaching Procedure.

After the results of the sediment toxicity assay were compiled, five sediment samples were chosen (sample 3, 5, 9, 17, and 18) to be subjected to an ATCLP. The selected samples represent some of the highest and lowest sediment toxicity results coupled with diverse sediment type and the Narraganset control sample (samples 3, 5, 9, 17, and control).

The ATCLP was adapted for use with daphnia acute toxicity tests by substituting CO<sub>2</sub>-saturated water for acetic acid. Carbonated water is most suitable for daphnia assays, because pH adjustment of the resultant extracts is seldom necessary.

Each sediment sample was sub-sampled and weighed into a pre-weighed 1-L EP tox jar. Saturated carbonic acid solution (extracting solution) was added to the sediment at a ratio equal to four times the mass of the sediment. The samples were agitated in the dark for 48 hr at 30 rpm (rotary end-over-end), and at a mean laboratory temperature of 20±2 °C. After agitation was completed, the soil was allowed to settle. The supernatant was decanted and filtered through a 0.45 μ filter and placed in a pre-cleaned EP tox jar and stored at 10 °C.

### 2.4 Daphnia Assays.

*Daphnia magna* were obtained from Dr. Freida Taub<sup>5</sup> (University of Washington, Seattle, WA). *Daphnia* were reared in the laboratory as described by Goulden, et al.<sup>6,7</sup> Stock cultures were fed a mixture of vitamin enriched unicellular green algae (*Ankistrodesmus falcatus*, *Selenastrum capricornutum* and *Chlamydomonas reinhardi*). *Daphnia* culture media was derived from well water passed through a limestone pH adjustment tank, a Zeata Sol iron removal system, carbon filtration and finally through a UV Sterilization system.

Ten *daphnia* <24 hr old were placed into 250 mL glass beakers filled with 100 mL of diluted extract solution. The extract was diluted to provide a concentration range from 100% to 6.2% volume to volume (dilution series were a factor of 0.5 apart). The test beakers were placed in a temperature controlled room (20 ± 2 °C) with a light/dark cycle of 16/8 hr with 65 ft candles of light. Two replicates were used in each test. Mortality was determined at 24 and 48 hr. If *daphnia* were not mobile, they were gently touched with a Pasteur pipette. Immobilization (mortality) was recorded if the *daphnia* could not swim actively for 15 s. The EC<sub>50</sub> (the effective concentration at which 50% of the organisms were immobilized) values were computed using the Probit analysis as prepared by Kessler.\* The EC<sub>50</sub> was also tabulated graphically, using a least square analysis to verify all Probit analyses.

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\*Florence, Kessler, U.S. Environmental Protection Agency, Cincinnati, OH, provided a statistical package for this study.

## 2.5

### MICROTOX Assay.

The MICROTOX assay utilizes a luminescent bacterium, *Photobacterium phosphoreum*. Toxicity determinations are based on an inversely proportional reduction of light output from the bacteria in response to toxic materials (as toxicity increases light output decreases). This assay is short (15 min) and can be conducted in the field if needed.

The MICROTOX 100% liquid-phase assay was conducted on the same sediment leachates samples used in the daphnia assays. The choice of the liquid-phase 100% assay was based on the ability to screen for samples of probable low toxicity, yet the test includes a percent sample concentration range of approximately 11-91%, with provision of an EC<sub>50</sub> calculation. Depending on the results derived from this screening assay, a more detailed extended assay is an available option.

Materials used in the MICROTOX assays were supplied by Microbics, Inc.,<sup>8</sup> which included lyophilized *P. phosphoreum* at approximately one hundred million cells per vial (reagent), 2% sodium chloride solution (diluent), and 22% sodium chloride solution (osmotic adjustment solution) for adjusting the osmotic pressure of concentrated samples not requiring pre-dilution with diluent as the stock.

Typically, the 100% assay is conducted within the temperature controlled wells of a photometer that consists of four sample dilutions (usually diluted by a factor of two) and one blank or control. Reagent is added directly to sample dilutions. All dilutions are prepared to have a final NaCl concentration of 2%, which is the natural environment of the microorganism. Following 5 and 15 min, readings are taken to measure any decrease in light output. The blank serves to correct the readings for time-dependent drift in light production. Light lost to light remaining is calculated and an EC<sub>50</sub> determined.

## 3. RESULTS

The sediments provided by Roy F. Weston, Incorporated, were classified as sand or mud, based on grain size analysis. Using the Wentworth grain size scale,<sup>9</sup> (Appendix B) sediments with 50% of the sample having grain sizes ranging from 2.0 - 0.0625 mm, was considered sand. Sediment with 50% of the grain size less than 0.0625 mm was considered mud (Table 2).

The toxicity of the sediment to the amphipods ranged from 21-98% mortality, with the Fairlee Creek and Rhode Island Controls being 26% and 6%, respectively (Table 3). Approximately 8 of 17 samples caused amphipod mortality above 50% (Figure 3). Of the eight samples with toxicity above 50%, six were sand and two were mud. Seven out of ten sediments that exhibited amphipod mortality <50% were sand.

The salinity of the sea water remained at 30 ppt throughout the testing period with a pH range of 7.4 - 8.1. The dissolved oxygen ranged from 7.9 - 8.2.

Several of the sediment samples were chosen to be subjected to daphnia and MICROTOX assays. The selection was based on the highest, lowest, and mid range toxicity to amphipods. The sediment samples selected were 3, 5, 9, 17 and 18. These samples were subjected to ATCLP to produce a leachate that was used in daphnia and MICROTOX assays. With the exception of sample 18 (the Rhode Island control), the leachates were not toxic to daphnia or the bacteria *P. phosphoreum* (Table 3). The Rhode Island control was slightly toxic to daphnia ( $EC_{50} = 77\%$ ) but not to *P. phosphoreum*.

The analysis using the MM-1 did not detect any hazardous volatile or semi-volatile materials in the sediments. The chromatograms obtained were very similar to each other, only showing various hydrocarbons that are inherently present within many sediment ("muck") samples.<sup>10,11</sup> The mass spectra of these compounds were indicative of hydrocarbon spectra, in that there was a continual loss of 14 atomic mass units (amu) between the mass fragmenting peaks. This is common of methylene ( $CH_2$ ) groups fragmenting off an alkane chain. Also, the signal/noise ratio of the alkanes/alkenes to the background noise of the instrument was small, indicating that these compounds were not volatile.

There were no mustard or nerve agents detected using head space analysis techniques. The detailed chemical screening (see Appendix C for the analyzed compounds, Roy F. Weston, Incorporated), did not show any detectable organics. However, there was a large variety of metals found, with sodium and calcium having the largest variance between samples (Table 4). When concentration of sodium, calcium, nitrogen, total organic carbon, and percent solids are presented in graphic form versus amphipod mortality (Figure 4, 5, 6, 7, and 8 respectively), some mortality trends can be seen. Amphipod mortality is inversely proportional to the concentration of sodium, calcium, nitrogen and total organic carbon. The amphipod mortality is proportional to the percent solids in the sediment. However, sediment from locations 7 and 13 do not fit these trends.

The complete metal analysis for samples number 3, 5, 7, 9, and 17 are listed in Table 4. Only the samples that were subjected to daphnia and MICROTOX assays are listed in order to give the reader more insight as to why some sediments were more toxic. A previously published report documents the metals detected in the remainder of the samples.<sup>12</sup>

#### 4. DISCUSSION

There is no doubt that the J-field toxic burning pits have various levels of terrestrial contamination, which is documented in the environmental studies conducted from the late 1970s to early 1980s. The major concern at the present time is whether the contamination at the site is effecting the surrounding areas.

Through wind, rain water run-off, and ground water seepage, there exist the possibility that contamination could effect the areas surrounding the disposal site. However, the area has not been disturbed for many years

and ground vegetation is plentiful. Therefore, contamination from wind and run-off should be minimal, if at all.

The sediment from the water ways surrounding the J-field disposal site typically have clay and organic debris, which have high affinity to organics and metals.<sup>13,14,15</sup> Sedentary sinks found in estuarine ecosystems, through sedimentation and chemical complexing, can collect heavy metals/organics and render them relatively non-toxic to pelagic organisms. However, the effects to bottom dwelling organisms may be impacted. Also, recreational water activities have potential for human dermal exposure to materials deposited in the sediments.

The sediment samples had a wide range of toxicity to amphipods, yet did not effect the daphnia or *P. phosphoreum* (leachate exposures). When the amphipods were collected from the wild, the consistency of the sediment was a black mud. Therefore, it was originally assumed that the toxicity associated with the sediments surrounding the J-Field disposal site was related to amphipod habitat preference. Many of the sediments taken from around the disposal site had a sandy consistency and did not seem to be suitable for amphipod habitat. Using the Wentworth grain size scale, Appendix B (grain size analysis were provided by Roy s. Weston, Incorporated), sediments with more than 50% of the sample having grain sizes ranging from 2.0 - 0.0625 mm was classified as sand. Sediment with 50% of the grain size <0.0625 mm was classified as mud (Table 2). The majority of the samples (6 out of 10) with <50% mortality to amphipods were classified as sand (Figure 3). Therefore, in this short term study, sediment preference can not be correlated to amphipod mortality.

There were a variety of metals detected in the sediments. Because there was no background metal analysis conducted before J-Field activities were started, determining the amount of naturally occurring metals in the sediments is difficult. The average concentrations of metals in the earth's crust are listed in Table 5.<sup>13</sup> However, using these figures in Table 5 to determine background metals found in the J-Field area may be erroneous, because these figures are world averages and some geographical areas may have much higher or lower natural concentrations. If the naturally occurring metal concentration for the east coast of the United States could be determined, then these concentration would be more justifiable to compare with the J-Field samples.

Schropp and Windom<sup>16,17</sup> have conducted research for estimating if metals in sediments are naturally occurring or contaminates. The ratio of naturally occurring heavy metals in sediments has been related to the concentration of aluminum contained in the sediment. Through log transformation of the aluminum/heavy metal ratio, regression plots can estimate if the sediments have been enriched (contain concentrations higher than background, contaminated). Using "clean" sediments from Florida estuarine areas, Schropp and Windom<sup>18</sup> have formulated regression plots for cadmium, arsenic, chromium, copper, zinc, nickel, and lead that can be used to estimate sediment contamination.

The J-Field data were subjected to the regression plots of Schropp and Windom assuming that naturally occurring metal ratios of Florida estuarine conditions were similar to Maryland estuarine conditions. As seen in Figure 9, 10, and 11, chromium, arsenic, and copper concentrations found in the sediments surrounding the J-Field disposal site are considered naturally occurring. Nickel and lead (Figure 12 and 13) show a slight enrichment; however, this could be attributed to laboratory error. Cadmium and zinc (Figure 14 and 15) show contamination above naturally occurring background levels. The enriched cadmium levels may not be justifiable, because cadmium was only detected in 5 of the 16 samples sites around J-Field. The elevated zinc levels were consistent in all the samples; therefore, urban activities up stream are most likely the major contributing factor. The Fairlee Creek reference site (sample 17) showed enrichment with copper, nickel, lead, cadmium, and zinc. Because the mouth of Fairlee Creek is narrow and there is a large boating community up stream, the elevated levels of metal can be related to recreational activities. The metal content found in the sediments surrounding the J-Field site can not be related to past disposal activities.

Heavy metal toxicity was considered a possible factor that contributed to the wide range of toxicity to amphipods. As seen in Table 4, the metal concentrations detected in the sediment from location 5 are much lower than any of the other locations listed, yet the toxicity to amphipods was 98%. The toxicity of the sediment can not be correlated to the heavy metal content. However, there are correlation's seen by graphing the concentrations of sodium, calcium, nitrogen, total organic carbon, and percent solids versus amphipod mortality (Figure 4, 5, 6, 7, and 8, respectively). Two samples (7 and 13) did not fit the correlation seen above. Several reasons for this occurrence exists; the samples could have been cross contaminated during sampling, laboratory error and/or an unusual unknown source. Trends seen in Figure 4, 5, 6, 7, and 8 indicate that the sediment samples toxic to amphipods may be due to acclimation problems dealing with the fresh water sediments. The sediments surrounding the J-Field site are considered fresh water. This study placed salt water (30 ppt) from Narraganset on top of freshwater sediments. The interaction between materials contained in the sediment and the salt water may have created ionic shifts that the amphipods could not tolerate.

## 5. CONCLUSION

The sediment surrounding the J-Field disposal site proved to have a wide range of toxicity to amphipods. The average (excluding the Rhode Island control) mortality rate to amphipods was approximately 48%. There were no organics related to the disposal site detected in the sediments. There was a variety of heavy metals detected in the sediment; however, the amphipod mortality could not be correlated to metal contamination. Also, there is no evidence to show that the metal contamination came from past J-Field disposal activities.

When comparing the concentration of sodium, calcium, nitrogen, total organic carbon, and percent solids to amphipod mortality, that trends seem to indicate that acclimation problems to fresh water sediments caused the

toxicity. Also, there were possibly organics that were not looked for that may have caused synergistic effects.

The chemistry of mixture toxicology has very complex interactions. Trying to identify the reasons why the amphipod mortalities had such a wide range is difficult. The interaction between metals, organics, clay materials, and freshwater sediments with overlaying salt water must be thoroughly understood before an explanation can be formulated to determine why amphipod mortality had such a wide range.

There is no evidence that can link past operations at the J-Field disposal site to any adverse effects seen during the exposure of amphipods to the sediments surrounding the disposal area.

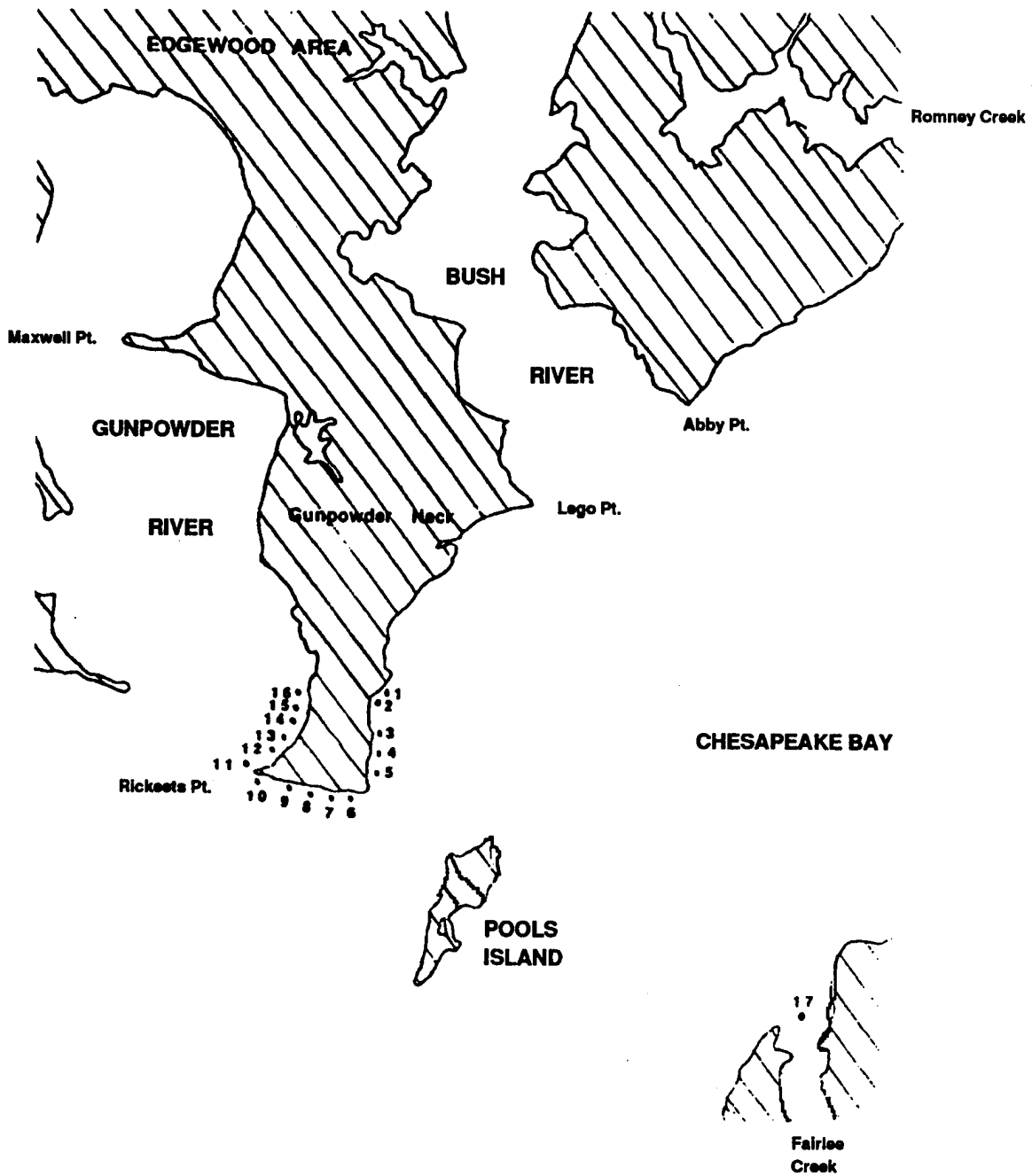


Figure 1. Map Location of Samples Taken Surrounding the J-Field Disposal Site

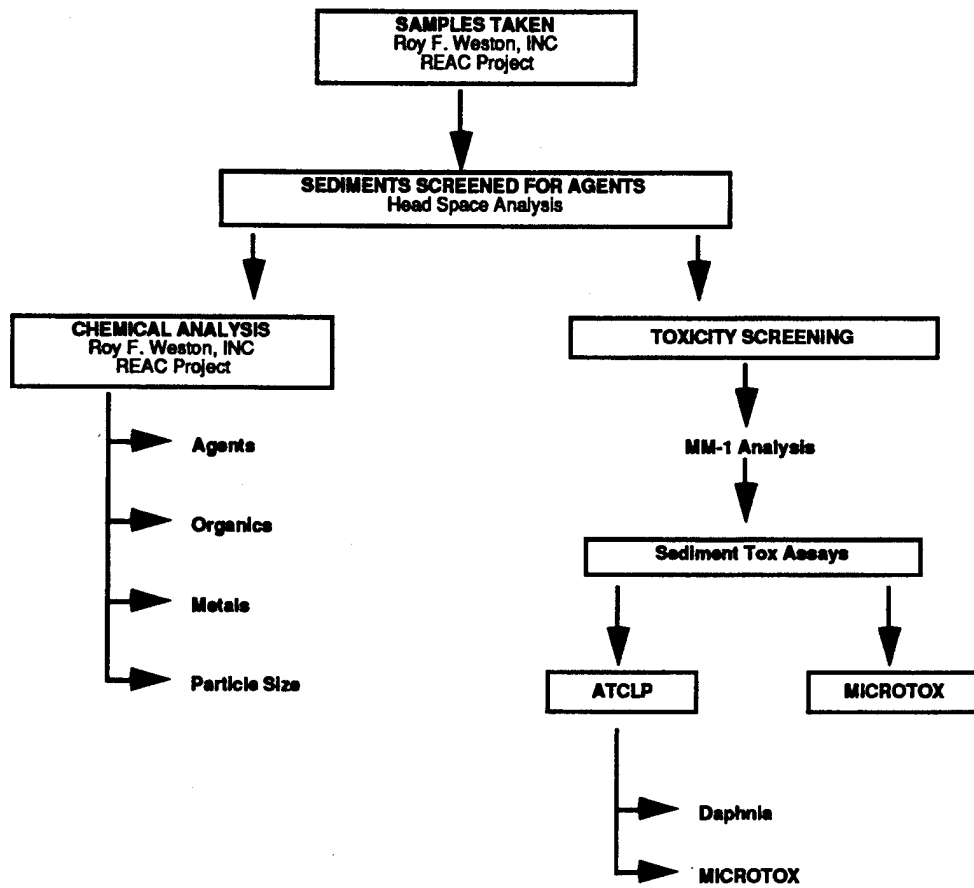


Figure 2. Flow Chart of Methods Used in Screening Sediment

### Amphipod Mortality

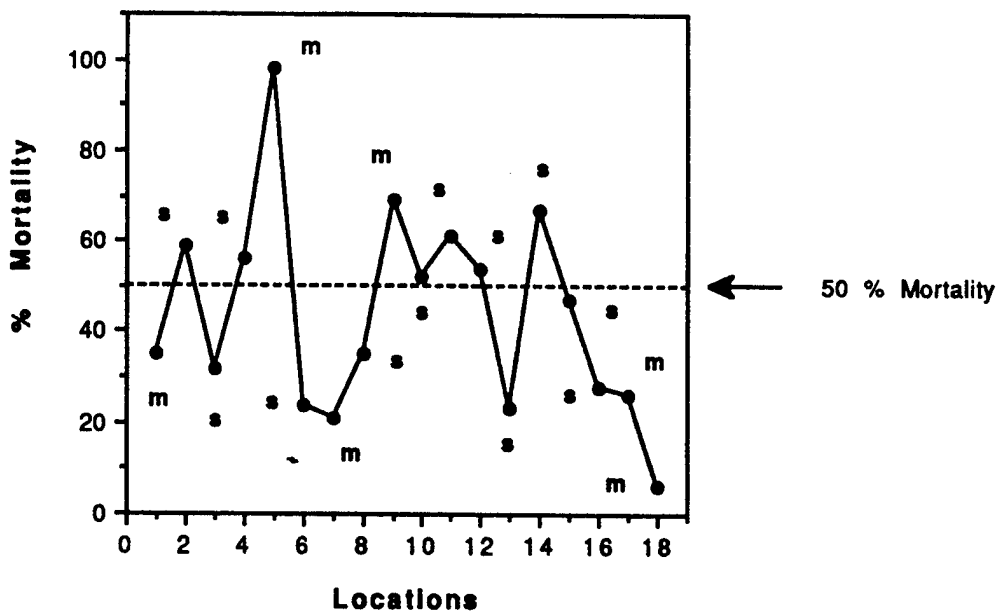


Figure 3. Amphipod Mortality Versus Sample Location. If sample contained <50% sand, the sediment was classified sand (s). If the sample contained >50% sand, the sediment was classified as mud (m).

### SODIUM

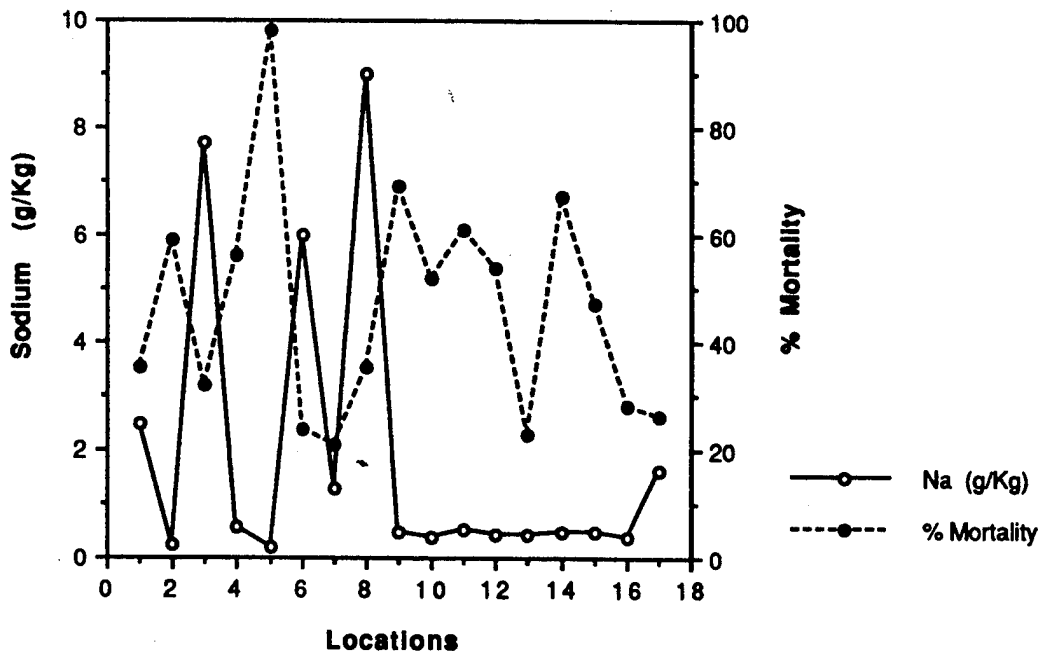


Figure 4. Concentration of Sodium in the Sediments Versus Mortality. Mortality is inversely proportional to the sodium concentration (sample 7 and 13 are exceptions).

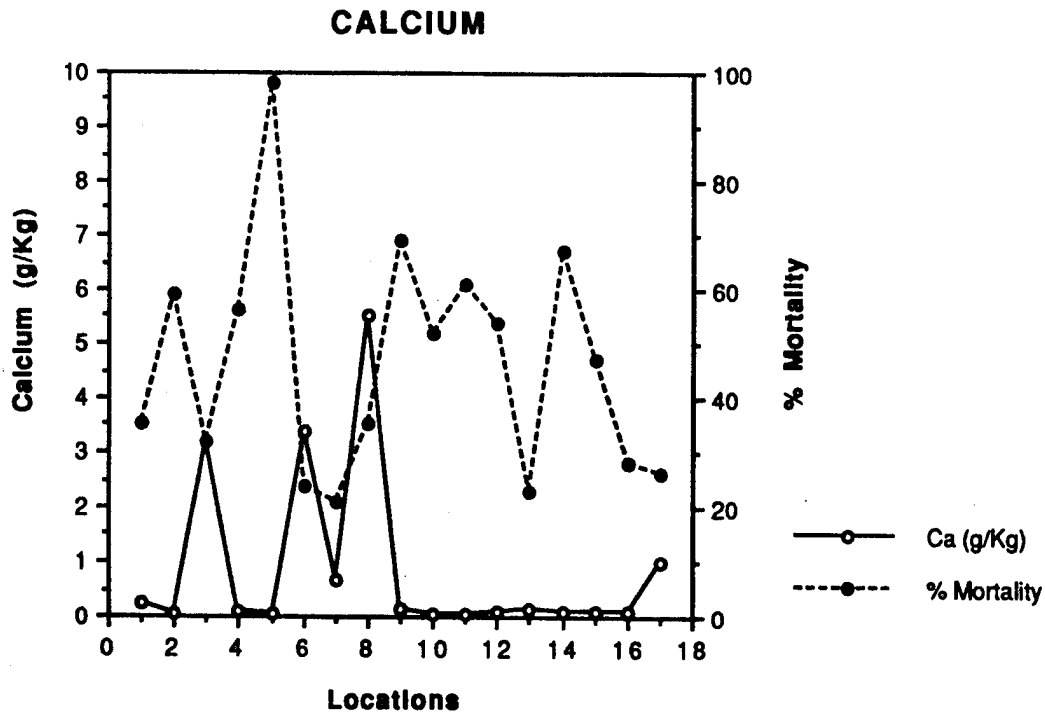


Figure 5. Concentration of Calcium in the Sediments Versus Mortality. Mortality is inversely proportional to the calcium concentration (sample 7 and 13 exceptions).

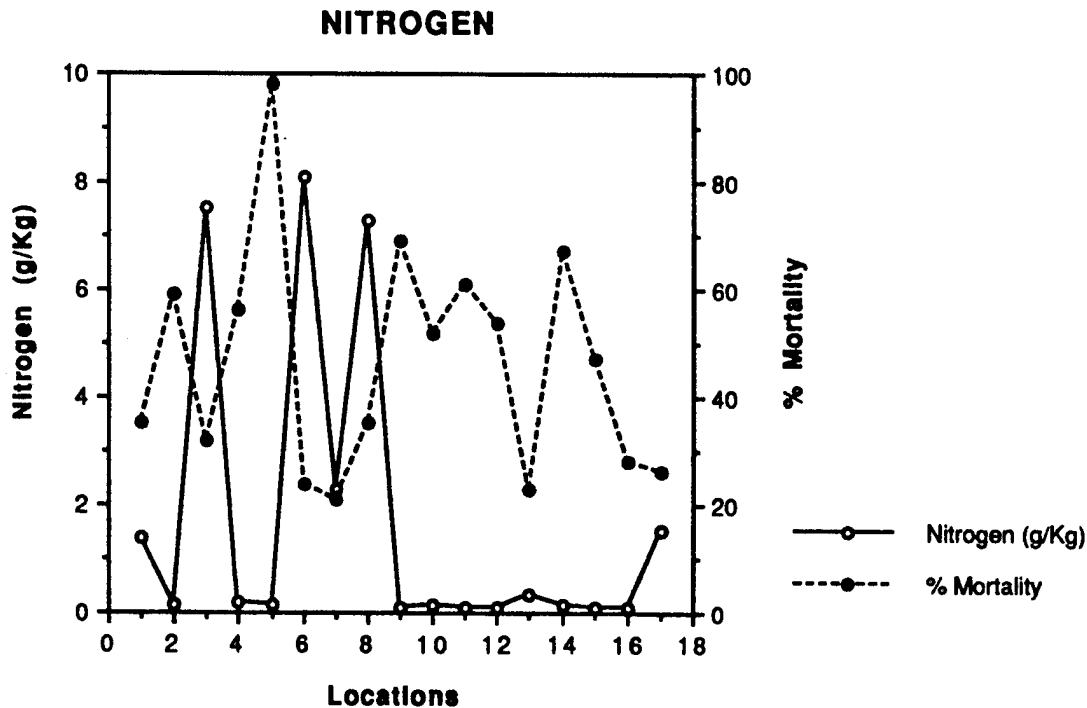


Figure 6. Concentration of Nitrogen in the Sediments Versus Mortality. Mortality is inversely proportional to the nitrogen concentration (sample 7 and 13 are exceptions).

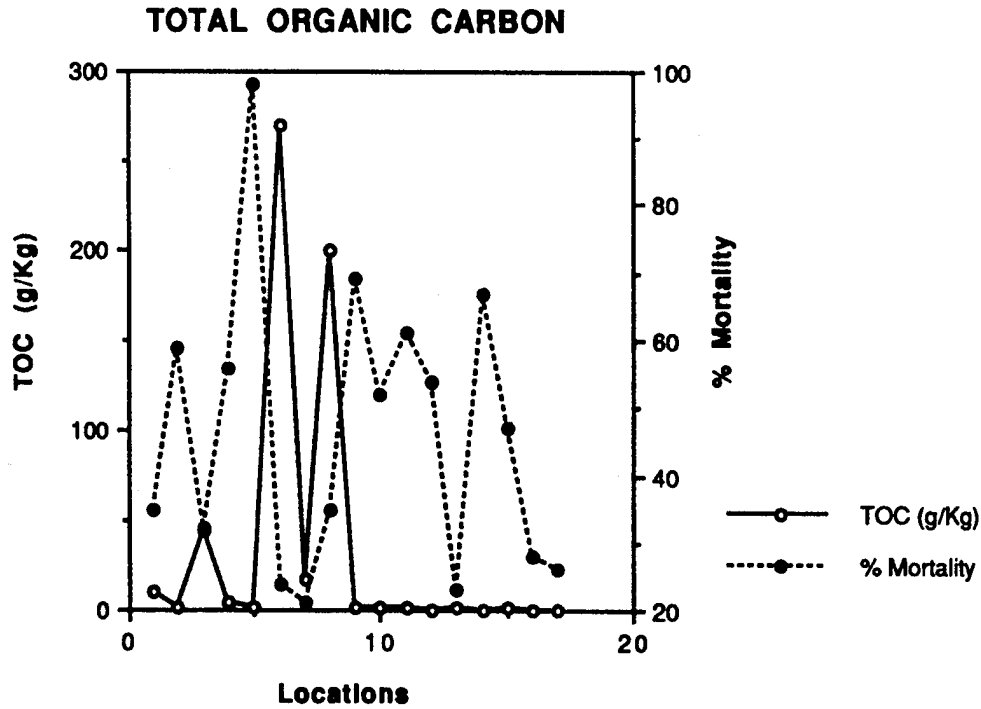


Figure 7. Total Organic Carbon in the Sediments Versus Mortality. Mortality is inversely proportional to the organic carbon concentration (sample 7 and 13 are exceptions).

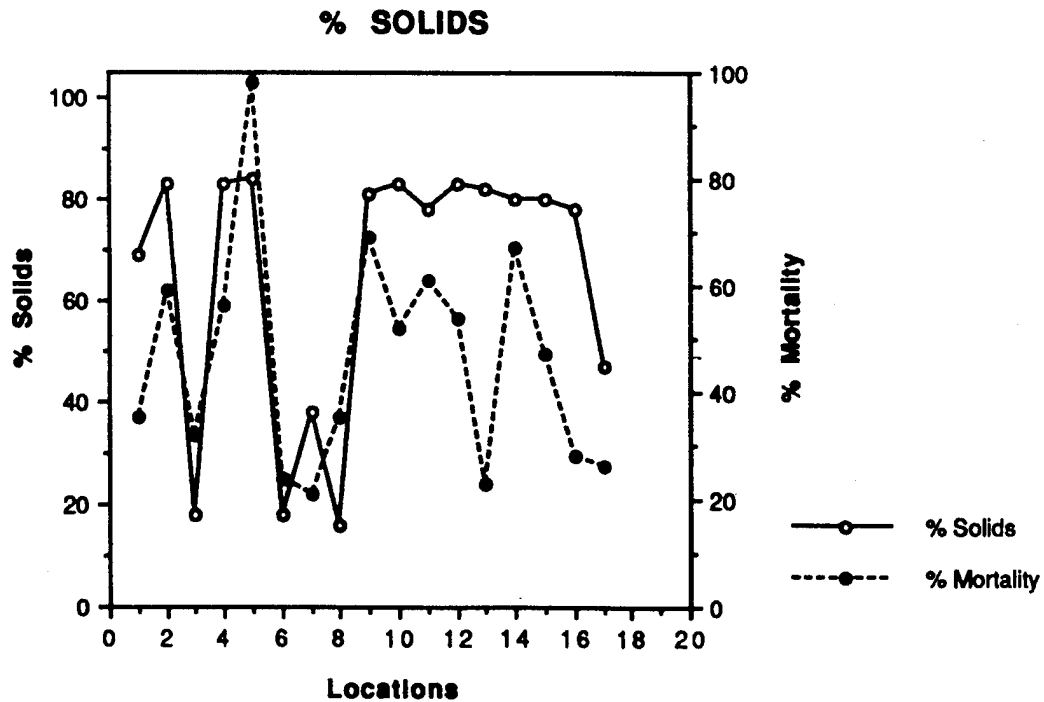
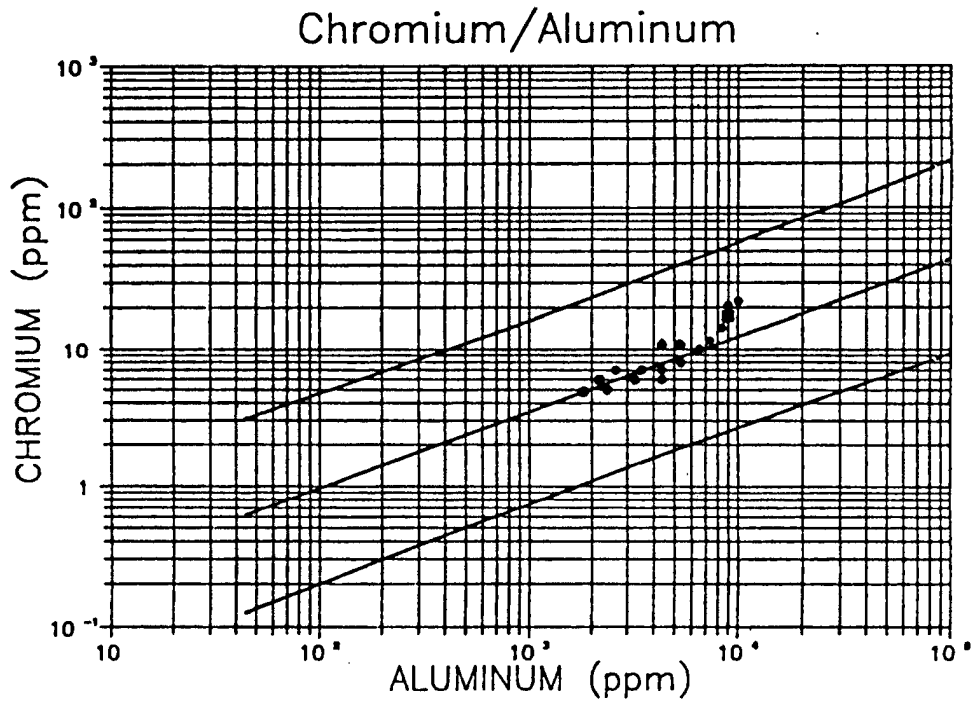


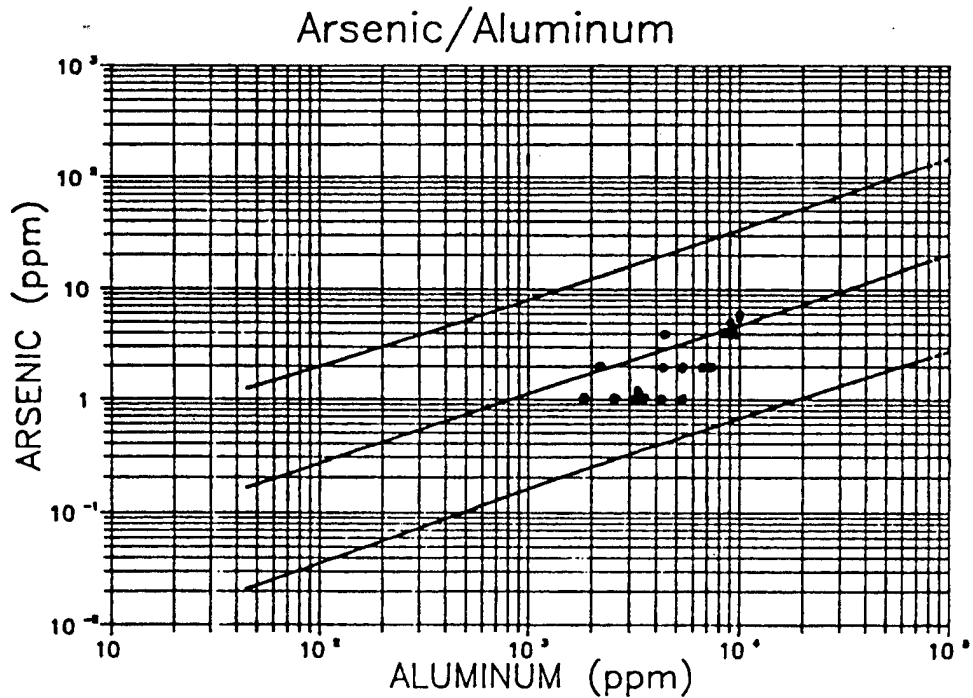
Figure 8. Percent Solids in the Sediments Versus Samples. As the percent solids increased the amphipod mortality increased (Samples 7 and 13 are exceptions)



■ Fairlee Creek Reference Site

Graphics template compliments of Schropp and Windom [22], State of Florida Department of Environmental Protection.

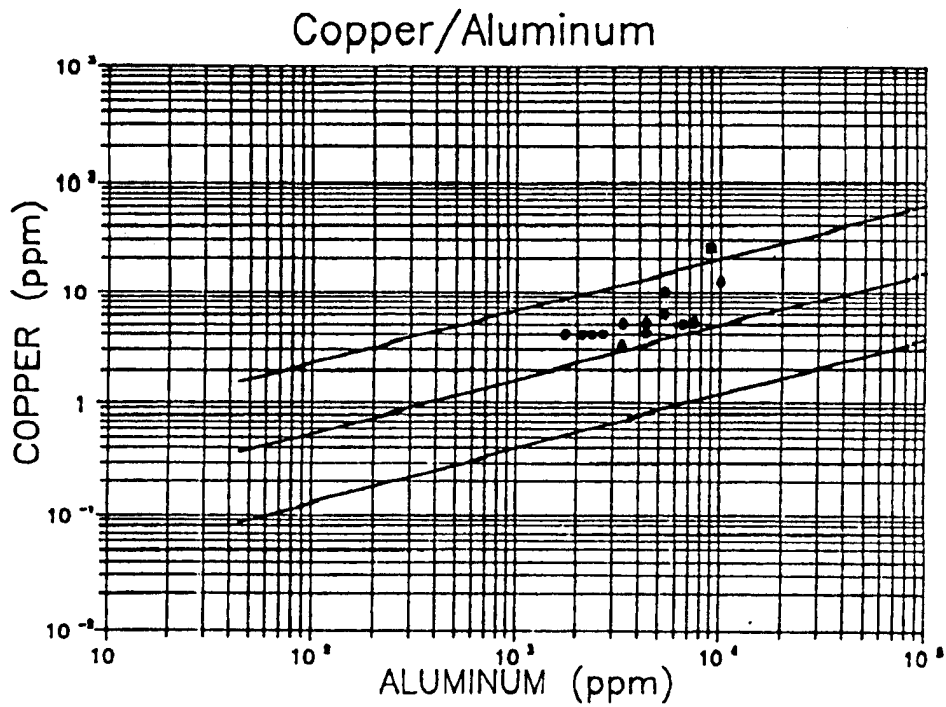
Figure 9. Chromium Concentrations Plotted Against Aluminum Found in Sediment Surrounding the J-Field Disposal Area. The regression line (center line) and the 95% confidence limits (upper and lower lines) were superimposed on the graph by Schropp and Windom. Points above the 95% limits are concidered enriched above back ground levels.



■ Fairlee Creek Reference Site

Graphics template compliments of Schropp and Windom [22], State of Florida Department of Environmental Protection.

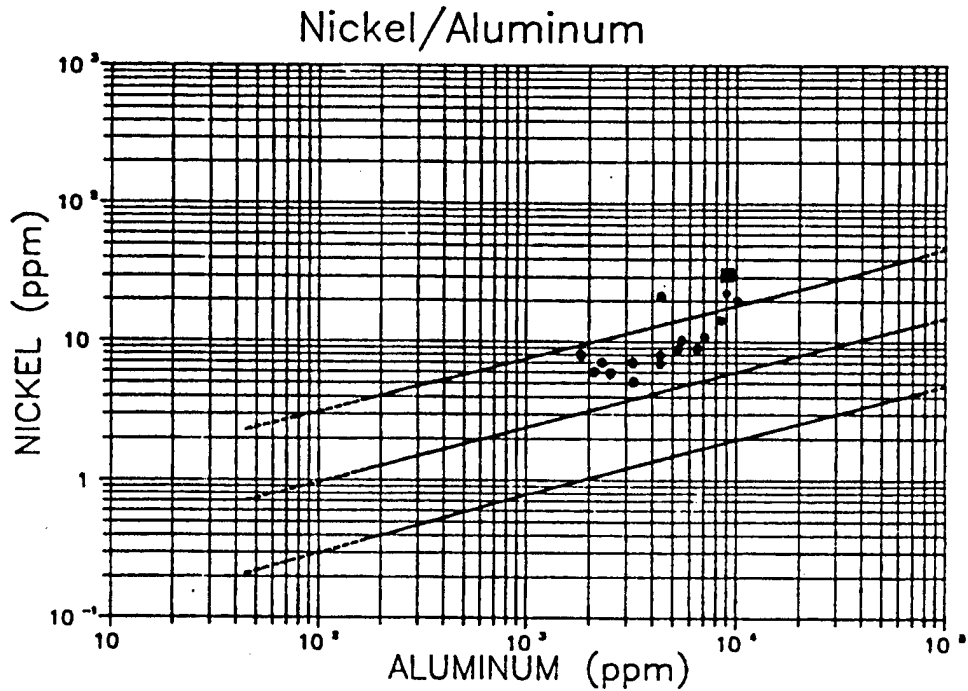
Figure 10. Arsenic Concentrations Plotted Against Aluminum Found in Sediment Surrounding the J-Field Disposal Area. The regression line (center line) and the 95% confidence limits were (upper and lower lines) superimposed on the graph by Schropp and Windom. Points above the 95% limits are concidered enriched above back ground levels.



■ Fairlee Creek Reference Site

Graphics template compliments of Schropp and Windom [22], State of Florida Department of Environmental Protection.

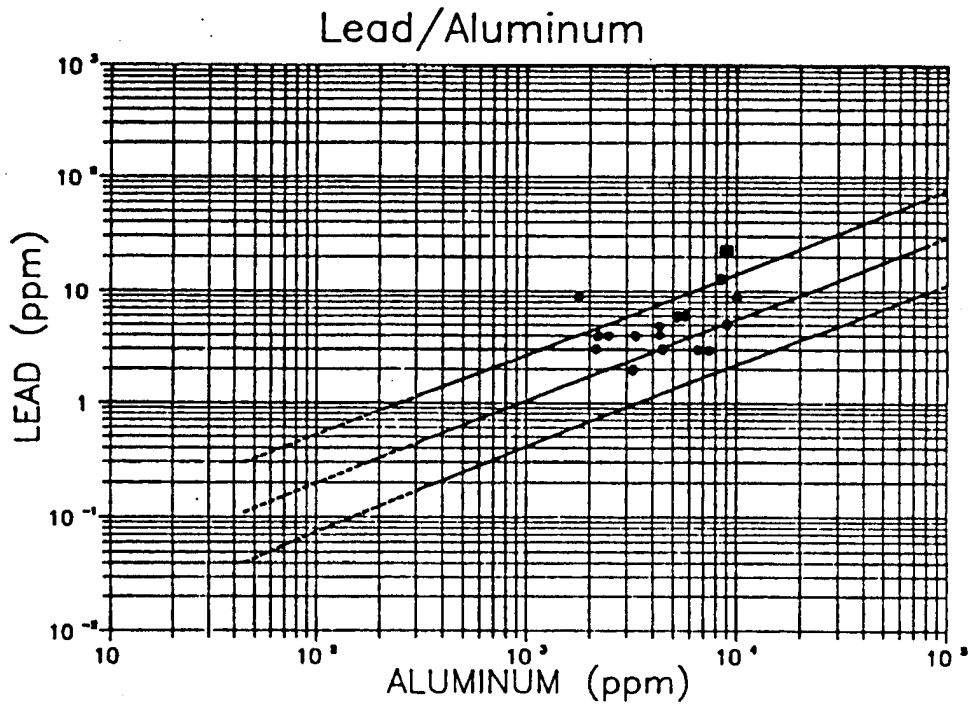
Figure 11. Copper Concentrations Plotted Against Aluminum Found in Sediment Surrounding the J-Field Disposal Area. The regression line (center line) and the 95% confidence limits (upper and lower lines) were superimposed on the graph by Schropp and Windom. Points above the 95% limits are concidered enriched above back ground levels.



■ Fairlee Creek Reference Site

Graphics template compliments of Schropp and Windom [22], State of Florida Department of Environmental Protection.

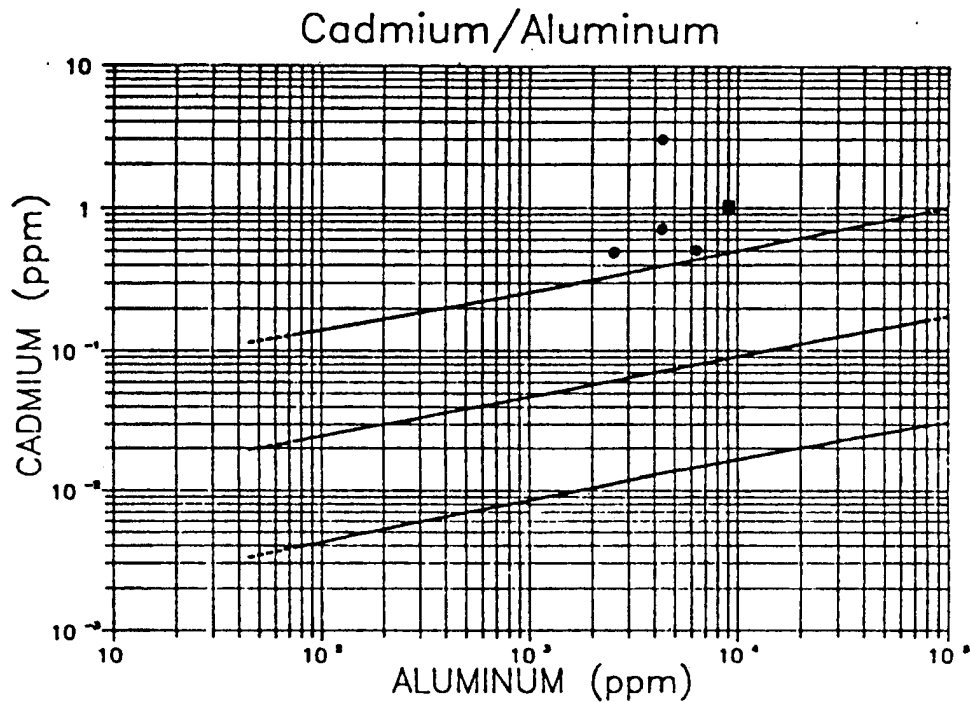
Figure 12. Nickel Concentrations Plotted Against Aluminum Found in Sediment Surrounding the J-Field Disposal Area. The regression line (center line) and the 95% confidence limits (upper and lower lines) were superimposed on the graph by Schropp and Windom. Points above the 95% limits are concidered enriched above back ground levels.



■ Fairlee Creek Reference Site

Graphics template compliments of Schropp and Windom [22], State of Florida Department of Environmental Protection.

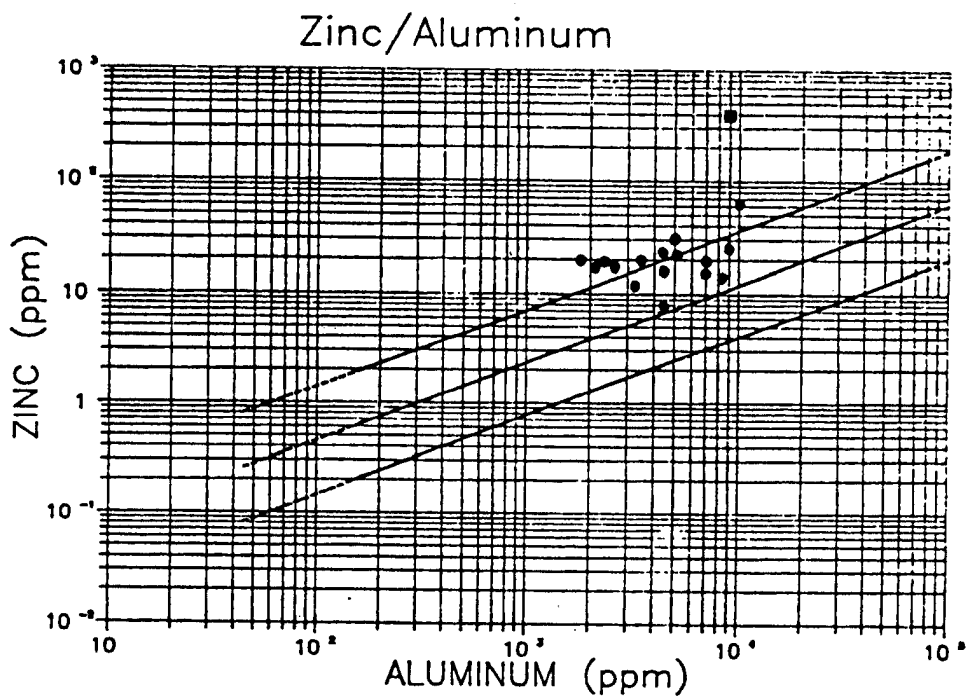
Figure 13. Lead Concentrations Plotted Against Aluminum Found in Sediment Surrounding the J-Field Disposal Area. The regression line (center line) and the 95% confidence limits (upper and lower lines) were superimposed on the graph by Schropp and Windom. Points above the 95% limits are concidered enriched above back ground levels.



■ Fairlee Creek Reference Site

Graphics template compliments of Schropp and Windom [22], State of Florida Department of Environmental Protection.

Figure 14. Cadmium Concentrations Plotted Against Aluminum Found in Sediment Surrounding the J-Field Disposal Area. The regression line (center line) and the 95% confidence limits (upper and lower lines) were superimposed on the graph by Schropp and Windom. Points above the 95% limits are concidered enriched above back ground levels.



■ Fairlee Creek Reference Site

Graphics template compliments of Schropp and Windom [22], State of Florida Department of Environmental Protection.

Figure 15. Zinc Concentrations Plotted Against Aluminum Found in Sediment Surrounding the J-Field Disposal Area. The regression line (center line) and the 95% confidence limits (upper and lower lines) were superimposed on the graph by Schropp and Windom. Points above the 95% limits are concidered enriched above back ground levels.

Table 1. Documented Materials Disposed at J-Field Disposal Site

Acronym	Chemical Name
GB	Isopropylmethylphosphonofluoride
GA	ethyl N,N-dimethylphosphoramino cyanidate
VX	O-ethyl S-(2-diisopropylaminoethyl) methyl phosphonothiolate
BZ	3-quinuclioinyl benzilate
H	bis(2-chloroethyl) sulfide
CNS	chloroacetophenone, chloroform and chloropicrin
CNC	chloroacetophenone in chloroform
CN	chloroacetophenone
DM	10-chloro-5,10-dihydrophenarsine
CS	2-chlorobenzal malononitrile
CG	Carbonyl chloride
WP	White Phosphorus
JP-8	Jet Fuel
Napalm	Gasoline, Benzene, and Polystyrene mix High explosives, Ignitors, Bursters

Table 2. Sediment Classification Based on the Wentworth Scale\*

Sample Location	% Sand	% Silt & Clay	Class
1	30.2	69.8	mud
2	67.9	32.1	sand
3	60.4	39.6	sand
4	27.5	72.5	mud
5	47.2	52.8	mud
6	66.2	33.8	sand
7	48.2	51.8	mud
8	78.6	21.4	sand
9	47.6	52.4	mud
10	84.6	15.4	sand
11	83.8	16.2	sand
12	83.3	16.7	sand
13	79.0	21.0	sand
14	81.4	18.6	sand
15	72.8	27.2	sand
16	90.5	9.5	sand
17	7.4	92.6	mud

\*Provided by Roy F. Weston, Incorporated, GSA Raritan, Depot, Edison, NJ.

Grain size analysis were grouped into % sands and % silt/clays. The sediment samples have been classified as mud (% sand <50) or (% sand >50).

Table 3. Toxicity Comparisons Between Test Organisms

Location #	<i>A. abdita</i> (Mortality)	<i>D. magna</i> (48 hr EC <sub>50</sub> )	<i>P. phosphoreum</i> (5 min & 15 min EC <sub>50</sub> )
1	35%		
2	59%		
3	32%	NT*	NT
4	56%		
5	98%	NT	NT
6	24%		
7	21%		
8	35%		
9	69%	NT	NT
10	52%		
11	61%		
12	54%		
13	23%		
14	67%		
15	47%		
16	28%		
17	26%	NT	NT
18	6%	77%**	NT

\*The leachate was not toxic to the organisms at 100%.

\*\*Dissolved oxygen at 100% leachate was 4.7%

Table 4. Metal Analysis from the Sediment Samples Used in the ATCLP

Metal (mg/kg)	Location 3	Location 5	Location 7	Location 9	Location 17
Aluminum	9000	5200	7200	5300	9000
Arsenic	5	1	2	2	4
Barium	23	19	17	23	47
Beryllium	BDL	BDL	0.7	BDL	BDL
Cadmium	BDL	BDL	BDL	BDL	1
Calcium	3200	24	680	150	1000
Chromium	20	8	11	11	19
Cobalt	BDL	5	7	6	20
Copper	BDL	6	5	10	26
Iron	14000	6100	9400	9000	20000
Lead	5	6	3	6	22
Magnesium	3100	200	400	180	1000
Manganese	210	36	53	50	470
Mercury	BDL	BDL	BDL	BDL	0.07
Nickel	24	9	11	10	31
Potassium	1600	320	550	340	1100
Sodium	7700	180	1300	480	1600
Vanadium	35	13	18	15	34
Zinc	27	27	18	30	130
Amphipod Mortality	32%	98%	21%	69%	26%

Metal analysis provided by Roy F. Weston, Incorporated, GSA Raritain, Depot, Edison, NJ.

Metals results from the remainder of the locations.<sup>16</sup>

Table 5. Average Metal Concentrations Found in the Earth's Crust\*

Metal	Earth Crust mg/kg
Aluminum	8.2 x 10 <sup>4</sup>
Arsenic	1.8
Barium	425.0
Beryllium	2.8
Cadmium	0.2
Calcium	4.2 x 10 <sup>4</sup>
Chromium	100.0
Cobalt	25.0
Copper	55.0
Iron	5.6 x 10 <sup>4</sup>
Lead	12.5
Magnesium	2.0 x 10 <sup>4</sup>
Manganese	940.0
Mercury	0.08
Nickel	75.0
Potassium	2.4 x 10 <sup>4</sup>
Sodium	2.4 x 10 <sup>4</sup>
Vanadium	135.0
Zinc	70.0

\*Figure taken from Riley, P.J. and Chester, R.,  
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## APPENDIX A

### CLEANSING PROCEDURE USED FOR CLEANING THE TEST CHAMBERS FOR THE SEDIMENT TOXICITY STUDY

The following is the glass ware washing methods used by Scientific Specialty Services (Randallstown, MD) for preparing pre-washed EP Tox Jars.

1. Washed in biodegradable non-phosphate soap.
2. Rinsed three times with tap water.
3. Rinsed with 1:1 nitric acid
4. Rinsed three times with deionized water.
5. Rinsed with hexane.
6. Oven dried.

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APPENDIX B

WENTWORTH GRAIN SIZE SCALE

The Wentworth Grain Size Scale for Sediments was used in classifying the sediment samples. For the purposes of interpretation the authors grouped sediment gradings into three classes sand, silt and clay. If the sand content was greater than 50%, the sediment was classified as sand. If the sand content was less than 50% the samples were classified as mud.

Description	Particle Size (mm)	Classification
Very Coarse Sand	2 - 1	Sands
Coarse Sand	1 - 0.5	
Medium Sand	0.5 - 0.25	
Fine Sand	0.25 - 0.125	
Very Fine Sand	0.125 - 0.0625	
Coarse Silt	0.0625 - 0.0312	Silts
Medium Silt	0.0312 - 0.0156	
Fine Silt	0.0156 - 0.00781	
Very Fine Silt	0.00781 - 0.00391	
Coarse Clay	0.00391 - 0.00195	Clays
Medium Clay	0.00195 - 0.0009765	
Fine Clay	0.0009765 - 0.0004882	
Very Fine Clay	< 0.0004882	

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## APPENDIX C

### CHEMICALS SCREENED FOR DURING THE CHEMICAL ANALYSIS OF THE SEDIMENTS

The following table lists the organics and metals analyzed for in the sediment samples. Chemical analysis provided by Roy F. Weston, Inc.

#### Agents

GA ethyl N,N-dimethyl phosphoraminothioate (nerve)  
GB isopropyl methyl phosphonofluoridate (nerve)  
GD pinacolyl methyl phosphonofluoridate (nerve)  
VX ethyl S-2-diisopropyl aminoethyl methylphosphorothioate(nerve)  
HD bis 2-chloroethyl sulfide (blister)  
HT (blister)  
BZ 3-quinuclidinyl benzilate(incapacitating)  
CN chloroacetophenone (tear)  
CS 2-chlorobenzal malononitrile(tear)

#### Organosulfur Degradation Products

Oxathiane  
1,4-Dithiane  
Benzothiane

#### Explosives

HMX Cyclotetramethylenetetranitramine  
RDX Cyclotrimethylenetrinitramine  
1,3-DNB 1,3-dinitrobenzene  
1,3,5-TNB 1,3,5-Trinitrobenzene  
NB Nitrobenzene  
Tetryl  
2,4,6-TNT 2,4,6-Trinitrotoluene  
2,4-DNT 2,4-Dinitrotoluene  
2,6-DNT 2,6-Dinitrotoluene  
2-NT 2-nitrotoluene  
3-NT 3-nitrotoluene  
4-NT 4-nitrotoluene

#### Organics

Chloromethane	Bromomethane
Vinyl Chloride	Chloroethane
Methylene Chloride	Acetone
Carbon Disulfide	1,1-Dichloroethene
1,1-dichloroethane	1,2-dichloroethylene
Chloroform	1,2-Dichloroethane
2-Butanone	1,1,1-Trichloroethane
Carbon Tetrachloride	Vinyl Acetate
Dichloropropane	Dichlorobromomethane
cis-1,3-Dichloropropylene	1,1,3-Trichloroethylene
Dibromochloromethane	1,1,2-trichloroethane
Benzene	trans-1,3-Dichloropropylene
Bromoform	4-Methyl-2-pentanone
2-Hexanone	2-Chloroethylvinylether
1,1,2,2-Tetrachloroethylene	1,1,2,2-Tetrachloroethane

## Organics

Phenol	bis-2-Chloroethylether
2-Chlorophenol	1,3-dichlorobenzene
1,3-Dichlorobenzene	Benzyl Alcohol
1,2-Dichlorobenzene	2-Methylphenol
bis-2-chloroisopropylether	4-Methylphenol
N-Nitroso-Di-n-propylamine	Hexachloroethane
Nitrobenzene	Isophorone
2-Nitrophenol	2,4-Dimethylphenol
1,2,4-Trichlorobenzene	Naphthalene
4-Chloroaniline	Hexachlorobutadiene
4-Chloro-3-methylphenol	2-Methylnaphthalene
Hexachlorocyclopentadiene	2,4,6-Trichlorophenol
2,4,5-Trichlorophenol	2-Chloronaphthalene
2-Nitroaniline	Dimethylphthalate
Acenaphthylene	3-Nitroaniline
Acenaphthene	2,4-Dinitrophenol
4-Nitrophenol	Dibenzofuran
Diethylphthalate	4-Chlorophenyl phenylether
Fluorene	4-Nitroaniline
4,6-Dinitro-2-methylphenol	N-Nitrosodiphenylamine
4-Bromophenol phenylether	Hexachlorobenzene
Pentachlorophenol	Phenanthrene
Anthracene	Carbazole
Di-n-butylphthalate	Fluoranthene
Pyrene	Butylbenzylphthalate
3,3-Dichlorobenzidine	Benzo(a)anthracene
bis-2-ethylhexyl phthalate	Chrysene
Di-n-octylphthalate	Benzo(b)fluoranthene
Benzo(k)fluoranthene	Benzo(a)pyrene
Indeno(1,2,3-cd)pyrene	Dibenzo(a,h)anthracene
Benzo(g,h,i)perylene	Chlorobenzene
Toluene	Ethyl Benzene
Styrene	Xylenes

## Metals

Aluminum	Antimony
Arsenic	Barium
Beryllium	Cadmium
Calcium	Chromium
Cobalt	Copper
Iron	Lead
Magnesium	Manganese
Mercury	Nickel
Potassium	Selenium
Silver	Sodium
Thallium	Vanadium
Zinc	

Pesticides

alpha-BHC	gamma-BHC
beta-BHC	Hepyachlor
delta-BHC	Aldrin
Heptachlor epoxide	gamma Chlordane
alpha Chlordane	Endosulfan
4,4-DDE	Dieldrin
Endrin	4,4-DDD
Endrin Aldehyde	Endosulfan sulfate
Methoxychlor	Endrin ketone
Toxaphene	PCBs