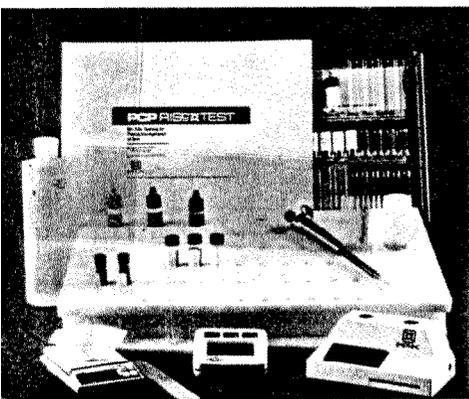
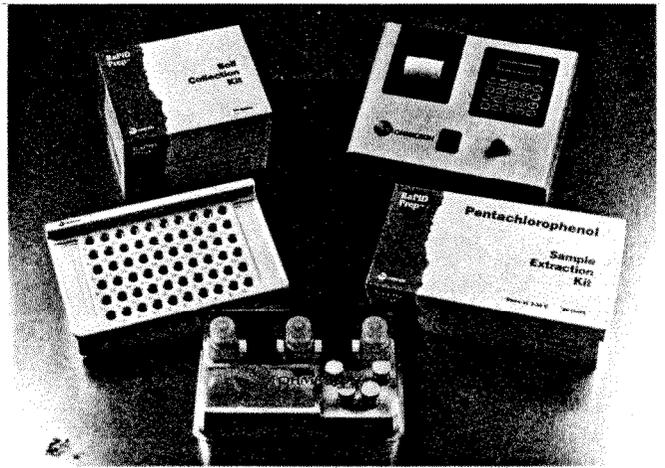
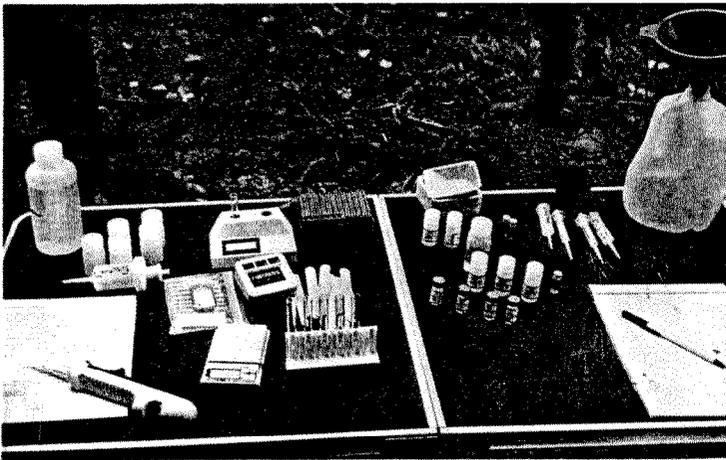




# PCP Immunoassay Technologies

## Innovative Technology Evaluation Report



SUPERFUND INNOVATIVE  
TECHNOLOGY EVALUATION

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# **PCP IMMUNOASSAY TECHNOLOGIES**

## **INNOVATIVE TECHNOLOGY EVALUATION REPORT**

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### **Notice**

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## Foreword

The U.S. Environmental Protection Agency is charged by Congress with protecting the Nation's land, air, and water resources. Under a mandate of national environmental laws, the Agency strives to formulate and implement actions leading to a compatible balance between human activities and the ability of natural systems to support and nurture life. To meet this mandate, EPA's research program is providing data and technical support for solving environmental problems today and building a science knowledge base necessary to manage our ecological resources wisely, understand how pollutants affect our health, and prevent or reduce environmental risks in the future.

The National Risk Management Research Laboratory is the Agency's center for investigation of technological and management approaches for reducing risks from threats to human health and the environment. The focus of the Laboratory's research program is on methods for the prevention and control of pollution to air, land, water and subsurface resources; protection of water quality in public water systems; remediation of contaminated sites and ground water; and prevention and control of indoor air pollution. The goal of this research effort is to catalyze development and implementation of innovative, cost-effective environmental technologies; develop scientific and engineering information needed by EPA to support regulatory and policy decisions; and provide technical support and information transfer to ensure effective implementation of environmental regulations and strategies.

This publication has been produced as part of the Laboratory's strategic long-term research plan. It is published and made available by EPA's Office of Research and Development to assist the user community and to link researchers with their clients.

E. Timothy Oppelt, Director  
National Risk Management Research Laboratory

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## **Abstract**

This report describes the demonstration and evaluation of three immunoassay field screening technologies designed to determine pentachlorophenol(PCP) contamination in soil and water. The three immunoassay technologies were (1) the Penta RISC Test System developed by EnSys, Inc., (2) the Penta RaPID Assay developed by Ohmicron Corporation, and (3) the EnviroGard Pentachlorophenol (PCP) Test Kit developed by Millipore Corporation. The technologies were demonstrated in Morrisville, North Carolina, in August 1993, by PRC Environmental Management, Inc. (PRC), under contract to the Environmental Protection Agency's (EPA) Environmental Monitoring Systems Laboratory-Las Vegas (EMSL-LV).

The principal objective of the demonstration was to evaluate each technology for accuracy and precision at detecting high and low levels of PCP in soil and water samples by comparing their results to those attained by a confirmatory laboratory using standard EPA analytical methods. Each technology also was qualitatively evaluated for the length of time required for analysis, ease of use, portability, and operating cost. Accuracy was also assessed through analysis of performance evaluation (PE) samples, and precision was further assessed by comparing the results obtained on duplicate samples. A secondary objective of the demonstration was to evaluate the specificity of each technology. The evaluation of specificity was performed by examining any problems due to naturally occurring matrix effects, site-specific matrix effects, and chemical cross reactivity. Information on specificity was gathered from each developer, from the analysis of demonstration samples, and from a specificity study performed during the demonstration.

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## List of Abbreviations and Acronyms

Beazer	Beazer East, Inc.
CCA	copper-chromium-arsenate
DCAA	2,4-dichlorophenylacetic acid
DQO	data quality objective
ECD	electron capture detector
ELISA	enzyme-linked immunosorbent assay
EMSL-LV	Environmental Monitoring Systems Laboratory-Las Vegas
EPA	Environmental Protection Agency
ERA	Environmental Research Associates
ESAT	Environmental Services Assistance Team
FASP	Field Analytical Screening Program
GC/MS	gas chromatograph/mass spectrograph
IDW	investigation-derived waste
ITER	Innovative Technology Evaluation Report
Koppers	Koppers Company
$\mu\text{g/L}$	micrograms per liter
$\mu\text{g/kg}$	micrograms per kilogram
MCL	maximum contaminant level
mg/kg	milligrams per kilogram
MMTP	Monitoring and Measurement Technologies Program
NRMRL	National Risk Management Research Laboratory
ORD	Office of Research and Development
OSWER	Office of Solid Waste and Emergency Response
PCP	pentachlorophenol
PE	performance evaluation
ppb	part per billion
ppm	part per million
PRC	PRC Environmental Management, Inc.
QADE	Quality Assurance and Data Evaluation
QA/QC	quality assurance/quality control
QAPjP	quality assurance project plan
RB	reagent blank
RCRA	Resource Conservation and Recovery Act
RECAP	Region 7 Environmental Collection and Analysis Program
RPD	relative percent difference
SARA	Superfund Amendments and Reauthorization Act of 1986
SITE	Superfund Innovative Technology Evaluation
SMO	Sample Management Office
SVOC	Semivolatile Organic Compound
US1	Unit Structures, Inc.

## **Acknowledgments**

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## Section 1 Executive Summary

This innovative technology evaluation report (ITER) presents information on the demonstration and evaluation of three immunoassay field screening technologies for determining pentachlorophenol (PCP) contamination in soil and water. These technologies were demonstrated in Morrisville, North Carolina, in August 1993. The demonstration was conducted by PRC Environmental Management, Inc. (PRC), under contract to the Environmental Protection Agency's (EPA) Environmental Monitoring Systems Laboratory-Las Vegas (EMSL-LV). The demonstration was developed under the Monitoring and Measurements Technologies Program (MMTP) of the Superfund Innovative Technology Evaluation (SITE) Program.

The three immunoassay technologies evaluated during this demonstration were (1) the Penta RISC Test System developed by EnSys, Inc., (2) the Penta RaPID Assay developed by Ohmicron Corporation, and (3) the EnviroGard Pentachlorophenol (PCP) Test Kit developed by Millipore Corporation. These technologies were demonstrated in conjunction with the demonstration of two other field screening technologies: the HNU-Hanby Test Kit developed by HNU Systems and the Field Analytical Screening Program (FASP) PCP Method developed by EPA's Region 7 under the Superfund Program. The demonstrations of these other two technologies are presented in separate reports similar to this one.

The first objective of this demonstration was to evaluate each of the field screening technologies for accuracy and precision in detecting high and low levels of PCP in soil and water samples by comparing their results to those attained by a confirmatory laboratory using standard EPA analytical methods. These EPA-approved methods are used to provide legally defensible analytical data for the purpose of monitoring or for the enforcement of environmental regulations. Because these EPA-approved methods are used by the regulatory community, this demonstration also used these methods. Though these methods may include inherent tendencies which may bias data or may include procedures with

which developers disagree, they are the best methods for providing legally defensible data as defined by the regulatory community. To remove as much of these inherent tendencies as possible, PRC used post-hoc residual analysis to remove data outliers. Each technology also was qualitatively evaluated for the length of time required for analysis, ease of use, portability, and operating cost.

The second objective of the demonstration was to evaluate the specificity of each technology. The evaluation of specificity was performed by examining any problems due to naturally occurring matrix effects, site-specific matrix effects, and chemical cross reactivity. Information on specificity was gathered from each developer, from the analysis of demonstration samples, and from a specificity study performed during the demonstration.

The site selected for demonstrating the technologies was the former Koppers Company (Koppers) site in Morrisville, North Carolina. One of the reasons this site was selected was because a National Risk Management Research Laboratory (NRMRL) SITE demonstration was occurring simultaneously, thus allowing EMSL-LV and NRMRL to combine logistical and support efforts. Another reason for selecting the former Koppers site was that historical documentation indicated its PCP contamination ranged from none detected to 3,200 parts per million (ppm) in soil and from none detected to 1,490 parts per billion (ppb) in groundwater. The PCP carrier used at this site was a mixture of isopropyl ether and butane. Soil and water samples also were collected from the Winona Post site in Winona, Missouri. These samples were shipped to the former Koppers site for inclusion as demonstration samples. Winona Post samples were included to broaden the scope of the demonstration by introducing a different sample matrix and PCP carrier, diesel fuel, to the evaluation of each technology. Findings for each of the immunoassay field screening technologies demonstrated are summarized below.

## **EnSys, Inc.: Penta RISc Test System**

The Penta RISc Test System is designed to quickly provide semiquantitative results for PCP concentrations in soil and water samples. In its standard format for soil analysis, the semiquantitative ranges assessed are: greater than 50 ppm, between 50 and 5 ppm, between 5 and 0.5 ppm, and less than 0.5 ppm. This technology's ranges for water analysis are greater than 5,000 ppb, between 5,000 and 500 ppb, between 500 and 5 ppb, and less than 5 ppb. The developer will customize these ranges to a user's needs. The immunoassay chemistry produces compound-specific reactions to PCP allowing its detection and quantitation. Polyclonal antibodies are fixed to the inside wall of a test tube where they offer binding sites for PCP. An enzyme conjugate containing a PCP derivative is added to the test tube to compete with PCP from samples for antibody binding sites. Excess sample and enzyme conjugate are removed from the test tube by washing, and reagents are added to the test tube to react with the enzyme conjugate causing a color formation. The amount of color formed by a sample is then compared to the color formed by a PCP standard taken through all of the immunoassay steps, simultaneously. The comparison is made with the use of a differential photometer.

The system is portable and can be operated outdoors; however, temperature extremes and humidity can affect its performance. It is easy to use even by those with no immunoassay testing experience. The highest number of demonstration samples analyzed in one 10-hour day was 40; the average number analyzed in one 10-hour day was 23. The detection limit reported by the developer for soil samples is 0.5 ppm and that for water samples is 5 ppb. The system can be affected by naturally occurring matrix effects such as humic acids, pH, or salinity. Site-specific matrix effects that can affect the system include PCP carriers, such as petroleum hydrocarbons or solvents, and other chemicals used in conjunction with PCP, such as creosote, copper-chromium-arsenate (CCA), or herbicides. Chemicals similar in structure to PCP can provide positive results. The system was found to be most affected by tetrachlorophenols and trichlorophenols. A specificity study performed during the demonstration showed that 2,3,4,6-tetrachlorophenol and 2,4,6-trichlorophenol would provide a positive response when present at a concentration of 10 ppm.

PRC evaluated field and laboratory duplicate samples to determine the technology's precision. The precision of the system for soil samples was found to be 79 percent. This is below the demonstration's criteria for acceptable precision. The precision of the system for water samples was found to be 100 percent. The comparison of the accuracy of the soil analysis showed that 73 percent of the time the technology was correct. The technology gave false positive results 19 percent of the time and gave false

negative results 8 percent of the time. All of the false negative results were for samples containing less than 10 ppm. When examined for a PCP carrier effect, the frequency of correct readings was higher for the samples collected at the Winona Post site. The frequency of false positive and false negative results was similar between the two sites. The system produced correct results 47 percent of the time for the water analysis. It had a 42 percent false positive rate and an 11 percent false negative rate. When this data was examined for a PCP carrier effect, no false negatives were reported on samples from the former Koppers site; in addition the frequency of correct and false positive results was higher for the samples collected from this site.

Overall, the technology was found not to be accurate when compared to Level 3 data, but this technology can produce Level 2 data. However, in some cases it produced only Level 1 data. The technology is conservative; however, using an absolute definition of accuracy, it was accurate only 73 percent of the time. A draft version of this ITER was distributed on March 4, 1994. EnSys submitted no comments on the draft ITER.

## **Ohmicron Corporation: Penta RaPID Assay**

The Penta RaPID Assay is designed to quickly provide quantitative results for PCP concentrations in soil and water samples. It uses immunoassay chemistry to produce compound-specific reactions to PCP allowing its detection and quantitation. Polyclonal antibodies are bound to paramagnetic particles and are introduced into a test tube where they offer binding sites for PCP. An enzyme conjugate containing a PCP derivative is added to the test tube where it competes with PCP from samples for antibody binding sites. A magnetic field is applied to each test tube to hold the antibodies containing the PCP and enzyme conjugate, while excess sample and enzyme conjugate are removed from the test tube by washing. Reagents are then added to the test tube where they react with the enzyme conjugate causing a color formation. A solution is added to each test tube to stop color formation. The color formed by a sample is then compared to the color formed by three PCP standards taken through all of the immunoassay steps. The comparison is made with the use of a spectrophotometer.

The technology is portable, but should be used indoors because fluctuations and extremes in temperature or humidity may affect its performance. Also, the reagents require refrigeration, and the spectrophotometer requires electricity. The technology was found to be easy to operate by individuals with some prior analytical laboratory experience, but can be used by individuals with no prior immunoassay testing experience. The highest number of samples analyzed in one 10-hour day during the demonstration was 64, and the average number of samples

analyzed in one 10-hour day was 21. The detection limit reported by the developer for soil samples is 0.1 ppm and for water samples is 0.06 ppb.

The technology may be affected by naturally occurring matrix effects such as humic acids, pH, or salinity. Site-specific matrix effects which can affect the technology include PCP carriers, such as petroleum hydrocarbons or solvents, other chemicals used in conjunction with PCP, such as creosote, CCA, or herbicides. Specific chemicals similar in structure to PCP can provide positive results with the technology. The technology was found to be most affected by tetrachlorophenols and trichlorophenols. A specificity study performed during the demonstration showed that 2,3,4,6-tetrachlorophenol and 2,4,6-trichlorophenol would provide a positive response when present at a concentration of 10 ppm.

PRC found no significant difference between the precision of the technology and that of the confirmatory laboratory. This conclusion was the same for both soil and water analysis. In addition, no PCP carrier effect on precision was observed. Results from the technology did not meet this demonstration's criteria for accuracy, as compared to Level 3 data. However, many of the data groupings produced were found to be linear, indicating that the results can be corrected mathematically. If 10 to 20 percent of the soil samples are sent to a confirmatory laboratory, then the results from the other 80 to 90 percent can be corrected. This need for mathematical correction to improve accuracy and the comparability of the technology's data with confirmatory data indicate that this technology can produce Level 2 analytical data. However, in some cases, this technology produced only Level 1 data. PRC found that the technology's water data was not significantly different from the confirmatory laboratory's when samples from the former Koppers site were analyzed. It indicated a significant difference between the data sets when the Winona Post site samples were analyzed. Therefore, the technology can produce Level 3 data for the former Koppers site water samples and Level 2 data for the Winona Post site water samples.

The developer submitted comments on a draft version of this report on May 4, 1994. These comments, which ranged from requests for clarification to technical comments on data interpretation, are available from EMSL-LV and PRC. In addition, the developer informed PRC that it now provides its customers with an "Environmental Users Guide," which advises them how to use the technology's quantitative data relative to action levels.

#### **Millipore Corporation: EnviroGard PCP Test Kit**

The EnviroGard PCP Test Kit is designed to quickly

provide semiquantitative or quantitative results for PCP concentrations in soil and water samples. Polyclonal antibodies are fixed to the inside wall of a test tube where they offer binding sites for PCP. An enzyme conjugate containing a PCP derivative is added to the test tube to compete with PCP from samples for antibody binding sites. Excess sample and enzyme conjugate is removed from the test tube by washing, and reagents are added to the test tube to react with the enzyme conjugate, causing color formation. The amount of color formed by a sample then is compared to the color formed by three PCP standards taken through all of the immunoassay steps. The results can be determined visually or a solution can be added to the test tube to stop color formation. A comparison of the sample and standards to a blank water sample is made with the use of a differential photometer. The differential photometer readings can be used to provide semiquantitative results or a standard curve can be prepared to allow for a quantitative determination.

The developer was extremely concerned about the results of this demonstration, particularly the results showing the technology's tendency to produce false negative results when concentrations of PCP were greater than 1,000 ppm. The developer began an investigation into the causes of this tendency and has revised the technology as a result. The modifications included the addition of a detergent wash instead of a water wash. About one third of the samples analyzed during the demonstration were then reanalyzed by the developer. The developer has said the false positive and false negative rates then both fell to 3 percent. The SITE Program, though, has not demonstrated and evaluated the technology as modified by the developer. All of the modifications have been incorporated into the technology's commercial product. The developer said in its letter to EPA regarding this demonstration: "We believe that, based on the knowledge gained from the field demonstration, we have been able to make significant improvements in our product. This experience demonstrates the utility of the SITE program and its goal of evaluating innovative technologies."

## Section 2 Introduction

This ITER summarizes the procedures used to demonstrate three immunoassay field screening technologies designed to detect PCP. The demonstration was conducted under the EPA's SITE Program by PRC. The three immunoassay technologies selected were: (1) the Penta RISC Test System developed by EnSys, Inc., (2) the Penta RaPID Assay developed by Ohmicron Corporation, and (3) the EnviroGard PCP Test Kit developed by the Millipore Corporation. These three immunoassay technologies were demonstrated in conjunction with the demonstration of two other screening technologies: the HNU-Hanby Test Kit developed by HNU Systems and the FASP PCP Method developed by EPA Region 7 under the Superfund Program. The results of the demonstration of these other technologies are presented in separate reports similar to this one.

### **EPA's Site Program and MMTP: An Overview**

At the time of the Superfund Amendments and Reauthorization Act of 1986 (SARA), it was well recognized that the environmental cleanup problem needed new and better methods. The SITE Program, therefore, was created to fulfill a requirement of SARA that the EPA address the potential of alternative or innovative technologies. The EPA made this program a joint effort between the Office of Solid Waste and Emergency Response (OSWER) and the Office of Research and Development (ORD). The SITE Program includes four parts:

- . The Demonstration Program (for remediation technologies)
- . The Emerging Technology Program
- . The Monitoring and Measurement Technologies Program (MMTP)
- . The Technology Transfer Program

The largest part of the SITE Program is concerned with treatment technologies and is administered by ORD's

NRMRL in Cincinnati, Ohio. The MMTP component, though, is administered by EMSL-LV. The MMTP is concerned with monitoring and measurement technologies that identify, quantify, or monitor changes in contaminants occurring at hazardous waste sites or that are used to characterize a site.

The MMTP seeks to identify and demonstrate innovative technologies that may provide less expensive, better, faster, or safer means of completing this monitoring or characterization. The managers of hazardous waste sites are often reluctant to use any method, other than conventional ones, to generate critical data on the nature and extent of contamination. It is generally understood that the courts recognize data generated with conventional laboratory methods; still, there is a tremendous need to generate data more cost effectively. Therefore, the EPA must identify innovative approaches, and through verifiable testing of the technologies under the SITE Program, ensure that the technologies are equivalent to or better than conventional technologies.

### ***The Role of Monitoring and Measurement Technologies***

Measurement and monitoring technologies are needed to assess the degree of contamination; to determine the effects of contamination on public health and the environment; to supply data for selection of appropriate remedial action; and to monitor the success or failure of selected remedies. Thus, the MMTP is concerned with evaluating screening technologies, including remote sensing, monitoring, and analytical technologies.

Candidate technologies may come from within the federal government or from the private sector. Through the program, developers are to rigorously evaluate the performance of their technologies. Finally, by distributing the results and recommendations of those evaluations, the market for the technologies is enhanced.

### ***Defining the Process***

The demonstration process begins by canvassing the

EPA's 10 regional offices (with input by OSWER and ORD) to determine their needs. Concurrently, classes of technologies are identified. An ideal match is made when there is a clear need by EPA's regions and a number of technologies that can address that need. The demonstrations are designed to judge each technology against existing standards and not "one against the other."

The demonstration is designed to provide for detailed quality assurance and quality control (QA/QC) to insure that a potential user can evaluate the accuracy, precision, representativeness, completeness, and comparability of data derived from the innovative technology. In addition, a description of the necessary steps and activities associated with operating the innovative technology is prepared. Cost data, critical to any environmental activity, are generated during the demonstration and allow a potential user to make economic comparisons. Finally, information on practical matters such as operator training requirements, detection levels, and ease of operation are reported. Thus, the demonstration report and other informational materials produced by MMTP provide a real-world comparison of that technology to traditional technologies. With cost and performance data, as well as "how to" information, users can determine whether a new technology better meets their needs.

### **Components of a Demonstration**

Once a decision has been made to demonstrate technologies to meet a particular EPA need, the MMTP performs a number of activities. First, the MMTP identifies potential participants and determines whether they are interested in participating. Each developer is advised of the general nature of the demonstration and is provided with information common to all MMTP demonstrations. Information is sought from each developer about its technology to insure that the technology meets the parameters of the demonstration. Then, after evaluation of the information, respondents are told whether they have been accepted into the demonstration or not. While participants are being identified, potential sites also are identified, and basic site information is obtained.

The next component, probably the most important, is the development of plans that describe how the demonstration will be conducted. A major part of the EPA's responsibility is the development of a demonstration plan, quality assurance project plan (QAPJP), and a health and safety plan. While the EPA pays for and has the primary responsibility for these plans, each is developed with input from all of the demonstration's participants. The plans define how activities will be conducted and how the technologies will be evaluated. The MMTP also provides each developer with site information and often predemonstration samples

so the developer can maximize the field performance of its innovative technology. Generally, the developers train EPA-designated personnel to operate their technologies so that performance is not based on the special expertise of the developers. This also insures that potential users have valid information on training requirements and the types of operators who typically use a technology successfully.

The field demonstration itself is the shortest part of the process. During the field demonstration, data is obtained on cost, technical effectiveness (compared to standard methods), and limiting factors. In addition, standardized field methods are developed and daily logs of activities and observations (including photos or videotape) are produced. The EPA is also responsible for the comparative, conventional method analytical costs and the disposal of any wastes generated by the field demonstration.

The final component of an MMTP demonstration consists of reporting the results and insuring distribution of demonstration information. The primary product of the demonstration is an ITER, like this one, which is peer-reviewed and distributed as part of the technology transfer responsibility of the MMTP. The ITER fully documents the procedures used during the field demonstration, QA/QC results, the field demonstration's results, and its conclusions. A separate QA/QC data package also is made available for those interested in evaluating the demonstration in greater depth. Two-page "Technical Briefs" are prepared to summarize the demonstration results and to insure rapid and wide distribution of the information.

Each developer is responsible for providing the equipment or technology product to be demonstrated, its own mobilization costs, and the training of EPA-designated operators. The MMTP does not provide any funds to developers for costs associated with preparation of equipment for demonstration or for development, and it does not cover the costs developers incur to demonstrate their products.

### **Rationale for this Demonstration**

PCP is a regulated chemical, is included in the EPA Extremely Hazardous Substances List, and is reported in the EPA Toxic Substances Control Act. Recently, PCP regulations under the Resource Conservation and Recovery Act (RCRA) have been created specifically for wood treatment facilities. PCP is included as a target compound of many EPA-approved analytical methods including: EPA 500 Series Methods 515.1 and 525, EPA 600 Series Methods 604 and 625, and EPA SW-846 Manual Methods 8040, 8151, 8250, and 8270. All of these methods use solvent extraction and gas chromatography. Detection and quantitation are

performed with flame ionization, electron capture, or mass spectrometer detectors. Analyzing samples for PCP using these methods is typically costly and time consuming. EMSL-LV identified the need for effective, accurate, low-cost screening technologies that could provide near real-time analytical data for PCP to Superfund and RCRA decisionmakers.

specificity, the length of time required for its analysis, ease of use, portability, and operating cost.

### **Demonstration Purpose, Goals, and Objectives**

Each of the three immunoassay technologies was evaluated on its accuracy and precision in detecting high and low levels of PCP in environmental samples, and the effects, if any, of both PCP carrier and natural matrix interferences on the technologies. The accuracy and precision of each technology were statistically compared to the accuracy and precision of a conventional confirmatory laboratory using EPA-approved analytical methods. These comparisons also were used to determine the highest data quality level that each technology could attain in field applications. For the purpose of this demonstration, the three primary data quality levels are defined as follows (EPA 1990):

- Level 1: This data is not necessarily compound-specific. Technologies that generate Level 1 data provide only an indication of contamination. Generally, the use of these technologies requires sample documentation, instrument calibration, and performance checks of equipment.
- Level 2: This data is compound-specific. To provide an accuracy check, verification analysis for at least 10 percent of the samples by an EPA-approved method is necessary. The method's analytical error is quantified. Use of QC procedures such as sample documentation, chain-of-custody procedures, sample holding time criteria, initial and continuing instrument calibration, method blank analysis, rinsate blank analysis, and trip blank analysis is recommended.
- Level 3: This data is considered formal or confirmatory analysis. Analytical error is quantified (precision, accuracy, coefficient of variation) and monitored. The following QC procedures are used: sample documentation, chains of custody, sample holding time criteria, initial and continuing instrument calibration, rinsate blank analysis, trip blank analysis, and PE samples. Detection limits are determined and monitored.

Each technology also was qualitatively evaluated for

## Section 3 Predemonstration Activities

Several activities were conducted by EMSL-LV, PRC, and other demonstration participants before the demonstration began. These activities included identifying developers, selecting the demonstration sites, selecting the confirmatory laboratory and analytical methods, conducting predemonstration sampling, and training technology operators. Predemonstration sampling and analysis are normally used to allow developers to refine their technologies and revise their operating instructions, if necessary, prior to the demonstration.

### Identifying Developers

EMSL-LV supplied PRC with the names of the immunoassay developers to be included in the demonstration, which were EnSys, Inc.; Ohmicron Corporation; and Millipore Corporation, and asked that PRC search for other technologies that could be included. The EPA Superfund Program's FASP PCP Method and the HNU-Hanby test kit produced by HNU Systems were included after PRC searched for other methods. These two methods are discussed in separate ITERs.

### Selecting the Sites

To evaluate the field screening technologies under field conditions, hazardous waste sites suitable for the demonstration were needed. The following criteria were used to select the appropriate sites:

- The technologies needed to be demonstrated at sites with a wide range of PCP contamination.
- PCP concentrations at the sites had to be well characterized and documented.
- The sites had to be accessible for conducting demonstration activities without interfering with any other activities being conducted on the site.

Because various carriers have been used with PCP and because those carriers may influence the technologies, it was determined that the sites used should offer two different carriers.

The former Koppers wood treatment site was selected as one of the two sites for this demonstration based on these criteria. This site also was selected because EPA's NRMRL was planning a SITE demonstration of the ETG Environmental, Inc., Base-Catalyzed Decomposition technology there and choosing the former Koppers site would allow logistical and support efforts between NRMRL and EMSLLV could be combined. The second demonstration site selected was the Winona Post site wood treatment facility. The Winona Post site is contaminated with PCP in a diesel fuel carrier solvent. The former Koppers site is contaminated with PCP in butane and isopropyl ether carrier solvents.

The former Koppers site is located in Morrisville, North Carolina, at the intersection of Highway 54 and Koppers Road. The site is currently owned by two companies: Beazer East, Inc. (Beazer), and Unit Structures, Inc. (USI). The portion of the site owned by Beazer is inactive. The portion of the site owned by USI is currently used as a wood laminating facility. The site occupies about 52 acres and includes the wood laminating building, an office, and several warehouses. Surrounding land use is a mixture of commercial, light industrial, and rural residential. During investigations at the former Koppers site, samples from the following media were collected: soil, groundwater, surface water, sediment, and fish. This sampling detected PCP from not detected to 3,200 ppm in soils and from not detected to 1,490 ppb in water.

The Winona Post site is located in Winona, Missouri, on Old Highway 60 West. It has operated as a saw milling and wood preserving facility since at least the early 1950s. The saw milling, wood preserving, and storage areas of the facility cover about 4 acres. The remaining portion of the 40-acre facility is wooded and largely undeveloped. The main features of the facility include a sawmill, office, treatment building, debarker, storage building, and pond. Currently, the company uses a solution of 5 percent PCP in diesel fuel. The solution is stored in the 20,000-gallon aboveground storage tank located adjacent to the treatment building. In the past, the Winona Post Company mixed its own solution

from concentrated PCP. Prior to the mid-1950s, the Winona Post Company treated wood with cresol. Six samples were collected at the Winona Post site in 1992 and revealed PCP concentrations ranging from 886 to 24,000 ppm in the soil and sediment samples and from 10 to 528 ppm in the surface water samples.

PCP is an organic chemical with an empirical formula of  $C_6Cl_5OH$  and a molecular weight of 266 grams per mole. PCP is an organic acid with a pKa of 4.7. PCP has a melting point of 191 °C and a boiling point of 310 °C. The specific gravity of PCP is 1.978 grams per cubic centimeter. PCP is described as slightly soluble in water, with 8 milligrams able to dissolve into 100 milliliters of water. The octanol-water partition coefficient of PCP is 6,400, which indicates that PCP is tightly bound to the soil matrix when it is released into the environment. PCP is used as a wood preservative, an insecticide, a preharvest defoliant, a slimicide, and a defoaming agent. The largest user of PCP is the wood treating industry. For treating wood, PCP is usually diluted to a 5 percent solution with solvents such as mineral spirits, kerosene, diesel fuel, or fuel oil. PCP also has been applied to wood with methylene chloride and liquified petroleum gas, such as butane. It has been manufactured under numerous trade names.

### **Selecting the Confirmatory Laboratory and Analytical Methods**

Before this demonstration, the EPA Region 7 Laboratory arranged for all soil samples to be analyzed under the Region 7 Environmental Collection and Analysis Program (RECAP) Contract and all water samples to be analyzed under its Environmental Services Assistance Team (ESAT) Contract. SW-846 protocols for Level 3 data were to be used to analyze soil and water samples during this demonstration. All samples were to be extracted by EPA Method 3540A and analyzed by EPA Method 8270A. Any soil samples in which PCP were not detected using Method 8270A were to be reanalyzed by Method 8151A calibrated to PCP. Use of Method 8151A would allow PCP to be detected at concentrations closer to the reported lower detection limits of the immunoassay technologies being demonstrated. Any groundwater samples in which PCP was not detected using Method 8270A were to be reanalyzed using Method 515.1. This method delivers detection levels for PCP in groundwater at or below the detection limits of the immunoassay technologies being evaluated. All of these analytical methods are well established and approved by EPA. The QA procedures, reporting requirements, and data quality objectives (DQO) of these methods are consistent with the goals of the SITE Program.

### **Training Technology Operators**

During the demonstration, the technologies were operated by PRC operators. Before the demonstration, these individuals were trained on how to use the technologies. This training involved a review of operating instructions provided by the developers and formal training by the developers. Training was equivalent to that recommended by the developers for users of their technologies on actual site characterization projects.

### **Predemonstration Sampling and Analysis**

In July 1993, PRC prepared a predemonstration sampling plan (PRC 1993a), and on July 12 1993, PRC collected predemonstration soil samples from areas at the former Koppers site previously identified as containing high, medium, low, and not detected concentrations of PCP. These samples were split into replicates. One replicate of each sample was submitted to each of the developers. These samples were not analyzed by a confirmatory laboratory because the contract for the confirmatory analyses were not yet finalized. The predemonstration sampling was limited to the former Koppers site. The Winona Post site was not added to the demonstration until after the predemonstration sampling had already occurred. All developers agreed to participate in the demonstration without having access to predemonstration samples from the Winona Post site. The unanimous finding from the developers regarding the predemonstration samples was that they did not exhibit their expected PCP concentrations.

## Section 4 Demonstration Design and Description

This section describes the organization of the demonstration, presents an overview of the demonstration's design, and details all deviations from the developer- and EPA-approved demonstration plan. Among the key portions of the demonstration plan presented here are the types of data collected and the statistical methods used to determine the accuracy and precision of the technologies. A detailed description of the demonstration is presented in the demonstration plan and QAPjP (PRC 1993b).

### Demonstration Design

The primary objective of the demonstration was to evaluate field portable analytical technologies for their effectiveness at detecting PCP in soil and water when operated in field conditions. This objective included defining the precision, accuracy, cost, and range of usefulness for each technology. A secondary objective was to define the DQOs that each technology could be used to address. The evaluation was designed so that the results from the technologies could be compared to those of a confirmatory laboratory that analyzed each sample using standard EPA-approved methods. The design limited, as much as possible, those elements of sample collection and analysis that would interfere with direct comparison of the results. These elements included heterogeneity of the samples and interference from other chemicals or other controllable sources.

The design also insured that the data was collected in a normal field environment. To do this, each technology was operated by an operator who worked in a trailer located at the former Koppers site. The operators were trained by representatives from each developer and were able to call the developers with questions when necessary. The operators, though, obtained all results on their own and reported the results once they believed the results were accurate and precise.

Standard QC samples were analyzed with each batch of environmental samples. Numerous laboratory and field duplicate samples were analyzed to insure a

proper measure of precision. The technologies were tested for common interferants. Qualitative measures, such as portability and ease of operation, were noted by the operators.

Overall, the demonstration was executed as planned in the demonstration plan and QAPjP. The final version of that plan was approved by all participants before the demonstration began. Below is a discussion of selected elements of that plan and a full discussion of deviations from it.

### Implementation of the Demonstration Plan

For the demonstration, 98 soil samples, 14 soil sample field duplicates, 10 water samples, and six water sample field duplicates were collected. Each soil sample was thoroughly homogenized and then split into replicate samples. One replicate from each water and soil sample was submitted to the confirmatory laboratory; three replicates were analyzed in trailers at the former Koppers site using the immunoassay technologies being evaluated. In addition to these samples, two soil PE samples and three water PE samples were analyzed by each technology.

The final demonstration plan called for the collection of 90 soil samples with the following distribution: (1) 40 samples containing 0 to 100 ppm PCP, (2) 25 samples containing 100 to 1,000 ppm PCP, and (3) 25 samples containing greater than 1,000 ppm PCP. During this demonstration, 98 soil samples were collected. The actual distribution of these samples, when the demonstration was complete, was as follows: (1) 60 samples contained 0 to 100 ppm PCP, (2) 16 samples contained 100 to 1,000 ppm PCP, and (3) 22 samples contained greater than 1,000 ppm PCP. This skewing of the sample set to the 0 to 100 ppm range should not affect the usability of this report since the majority of EPA PCP soil action levels occur in the 20 to 100 ppm range.

Of the samples collected for the demonstration, 53 soil samples, 9 soil field duplicates, 5 water samples, and 5 water field duplicate samples were collected at the former Koppers site. The soil samples were collected from areas known to exhibit a wide range of PCP concentrations. The areas sampled ranged in PCP concentration from not detected to 3,220 ppm. Most of the samples were collected from areas characterized during the remedial investigation. The water samples were collected from five existing groundwater monitoring wells located at the former Koppers site. The PCP concentrations in these wells were well documented from past sampling. The PCP concentrations sampled ranged from not detected to almost 1,500 ppb.

Of the samples collected for the demonstration, 45 soil samples, 5 soil field duplicates, 5 water samples, and 1 water duplicate sample were collected at the Winona Post site. The soil samples were collected in areas believed to be contaminated with high (greater than 1,000 ppm), medium (100 to 999 ppm), and low concentrations (less than 99 ppm) of PCP. The identification of these areas was based on past sampling data and visual signs of waste disposal. The water samples were collected from surface water located on or near the site. All of the Winona Post samples were collected, packaged, and shipped to the former Koppers site using the methods in the demonstration plan.

### **Field Modifications to the Demonstration Plan**

Two field modifications were made to the approved demonstration plan. First, fluorescein was not added to the soil samples prior to homogenization, as specified in the demonstration plan. The nature of the soil samples at both the former Koppers site and the Winona Post site allowed easy and thorough homogenization. The saturated stiff clay matrix for which the fluorescein additions were designed was not encountered at the former Koppers site, and thus, for consistency, this technique was eliminated at both sites. PRC believes that the elimination of the fluorescein from the homogenization process was offset by the long homogenization times used during this demonstration. To further examine this position, PRC conducted a side-by-side comparison of homogenization with and without fluorescent. Samples from the former Koppers site were used for this comparison. Due to the dry nature of the soil, the soil had to be hydrated with water to allow visible distribution of the fluorescein. The addition of the water and fluorescein caused a two-unit increase in the soil sample pH. This alteration of the sample chemistry coupled with the reactive nature of PCP invalidated the fluorescein homogenization

approach for environmental applications. PRC used an EPA-approved homogenization method and applied it to each sample for between 10 and 15 minutes. This method involved vigorous kneading of the sample in a clear plastic bag.

The second modification to the approved demonstration plan involved the sampling of the water matrix. The change was made because the EPA Region 7 project sponsor altered the design of the demonstration with regard to the evaluation of the water assays. The EPA project sponsor required that the number of water samples collected and analyzed for the demonstration be reduced to a total of 5 or 10. The approved demonstration plan called for the collection of 50 groundwater samples. To maximize the usefulness of the reduced number of water samples, PRC and EMSL-LV agreed to combine data from both sites if they could be shown statistically to come from the same distribution, thereby increasing the sample set size. Also, the EPA Region 7 project sponsor agreed to allow the following: (1) the Region 7 Laboratory would split and analyze samples from the Winona Post site in sample-plus duplicate (split) pairs; (2) the excess water from the original Winona Post site water sample that was duplicated would be used for laboratory QA/QC; and (3) only five monitoring wells would be sampled at the former Koppers site, and each sample would be duplicated. This would have resulted in five paired (sample plus its duplicate) samples from each site. This in turn would have provided five samples from each site for an accuracy assessment, and five paired samples from each site for a precision assessment. Although these are minimal sample sizes, it was felt that this design would provide the most useful data given the reduction in analytical resources that the EPA Region 7 sponsor required. However, when PRC delivered the soil samples and the water samples from the former Koppers site, it learned that the Winona Post site water samples had been extracted and analyzed as they were delivered, as five environmental samples with one duplicate. This failure to follow the modified experimental design resulted in only four single sample results and one duplicate result for precision analysis for this data set. The former Koppers site data set consisted of five duplicate pairs.

### **Data Collection**

The technology operators prepared a subjective evaluation of how difficult each technology was to use. Other qualitative measures included portability, ruggedness, instrument reliability, and health and safety considerations. Information on these qualitative factors was collected both by the operator of each technology and by the project's lead chemist.

Accuracy and precision were statistically evaluated during this demonstration. To evaluate accuracy and precision, all samples collected for the demonstration were split between the technologies and the confirmatory laboratory for analysis. The results from the confirmatory laboratory, for the purposes of this demonstration, were considered the actual concentration of PCP in each sample. The cost of using each technology also was assessed. Cost, for the purposes of this demonstration, included expendable supplies, nonexpendable equipment, labor, and investigation-derived waste (IDW) disposal. These costs were tracked during the demonstration.

### **Statistical Analysis of Results**

For each technology, two data sets were created: one for soil samples and the other for water samples. In addition, each water and soil data set was composed of two subsets, one for the samples taken from the former Koppers site and one for those collected at the Winona Post site. This grouping was intended to assess potential PCP carrier effects. A third data separation involved grouping the site-specific data sets into results greater than 100 ppm PCP and less than 100 ppm PCP. This grouping was intended to assess potential concentration effects on the data analysis.

These data sets were prepared for the statistical analysis following the methods detailed in the approved demonstration plan. When comparing duplicate samples or when comparing the results of a technology to those from the confirmatory laboratory, sample pairs that contained a nondetect were removed from the data sets. While other statistical methods can be used when nondetects are encountered, PRC felt that the variance introduced by eliminating these data pairs would be less than, or no more than equal to, the variance produced by giving these results an arbitrary value.

### **Intramethod Comparisons**

Sample results from each technology were compared to their duplicate sample results and to other QA/QC sample results. These comparisons are called intramethod comparisons. Intramethod accuracy was measured by assessing each technology's performance in analyzing PE samples. If the method produced a result considered accurate by the manufacturer that produced the PE samples, the technology was considered to have acceptable intramethod accuracy for this demonstration. Intramethod precision was assessed through the statistical analysis of relative percent differences (RPD). First, the RPDs of the results for each sample pair, in which both the sample and its duplicate were found to contain PCP,

were determined. The RPDs then were compared to upper and lower control limits. When using conventional technologies, such data is often available from analysis of samples collected during previous investigations. Because the technologies being demonstrated were themselves being assessed, the control limits used were calculated from data provided during this investigation. To determine these control limits, the standard deviation of the RPDs was calculated for each technology. This standard deviation was then multiplied by two and added to its respective mean RPDs. This established the upper control limit for the technology. Because an RPD of zero would mean that the duplicate samples matched their respective samples perfectly, zero was used as the lower control limit. This resulted in a large range of acceptable values. Because duplicate analyses seldom match perfectly, even for established technologies, all samples that fell within the control limits were considered acceptable. PRC determined that if at least 90 percent of the duplicate samples fell within these control limits, the technology had acceptable intramethod precision.

### **Intermethod Comparisons**

The data sets from the technologies, also, were statistically compared to the results from the confirmatory laboratory, and the precision of a technology was statistically compared to the precision of the confirmatory laboratory. These comparisons are called intermethod comparisons. In both cases, the results from the confirmatory laboratory were considered to be as accurate and precise as analytically possible.

The statistical methods used to determine intermethod accuracy were linear regression analysis, the Wilcoxon Signed Ranks Test, and the Fisher's Test. Linear regression was used for the technologies that were capable of determining quantitative results. PRC further prepared the data sets for the linear regression by averaging the field duplicate results. This was done to insure that samples were not unduly weighted in the regression analysis. This further preparation of the data sets was presented in the demonstration plan and agreed to by the developers before the demonstration began.

PRC calculated linear regression by the method of least squares. Calculating linear regression in this way makes it possible to determine whether two sets of data are reasonably related, and if so, how closely. Calculating linear regression results in an equation that can be visually expressed as a line. Three factors are determined during calculations of linear regression. These three factors are the y-intercept, the slope of the line, and the correlation coefficient, also called an  $r^2$ .

All three of these factors had to have acceptable values before a technology's accuracy was considered to meet Level 3 data quality requirements.

The  $r^2$  expresses the mathematical relationship between two data sets. If the  $r^2$  is one, then the two data sets are directly related. Lower  $r^2$  values indicate less of a relationship. Because of the heterogeneous nature of environmental samples,  $r^2$  values between 0.85 and 1 were considered to meet data quality Level 3 accuracy requirements;  $r^2$  values between 0.75 and 0.85 were considered to meet data quality Level 2 accuracy requirements; and  $r^2$  values below 0.75 were considered not accurate, meeting, at best, Level 1 accuracy requirements. The classification of data as Levels 2 or 1 was implied in the approved demonstration plan; however, these specific criteria were not presented.

If the regression analysis resulted in an  $r^2$  between 0.85 and 1, then the regression line's y-intercept and slope were examined to determine how closely the two data sets matched. A slope of one and a y-intercept of zero would mean that the results of the technology matched those of the confirmatory laboratory perfectly. Theoretically, the farther the slope and y-intercept differ from these expected values, the less accurate the technology. Still, a slope or y-intercept can differ slightly from their expected values without that difference being statistically significant. To determine whether such differences were statistically significant, PRC used the normal deviate test statistic. This test statistic results in a value that is compared to a table. The value at the 90 percent confidence level was used for the comparison. To meet data quality Level 3 requirements, both the slope and y-intercept had to be statistically the same as their ideal values.

If the  $r^2$  was between 0.75 and 1, and one or both of the other two regression parameters were not equal to their ideal, the technology was considered inaccurate but producing Level 2 quality data. Results in this case could be mathematically corrected if 10 to 20 percent of the samples were sent to a confirmatory laboratory. Analysis of a percentage of the samples by a confirmatory laboratory would provide a basis for determining a correction factor. Only in cases where the  $r^2$ , the y-intercept, and the slope were all found to be acceptable did PRC determine that the technology was accurate, meeting Level 3 data quality requirements.

Data placed in the Level 1 category had  $r^2$  values less than 0.75, the data was not statistically similar to the confirmatory data, based on parametric testing, or the results did not meet the manufacturer's performance specifications.

A second statistical method used to assess the intermethod accuracy of the data from each technology was the Wilcoxon Signed Ranks Test. This test is a nonparametric method for comparing matched pairs of data. It can be used to evaluate whether two sets of data are significantly different. The test requires no assumption regarding the population distribution of the two sets of data being evaluated other than that the distributions will occur identically. In other words, when one data point deviates, its respective point in the other set of data will deviate similarly. Because the only deviation expected during the demonstration was a difference in the concentrations reported by each technology, the two sets of data were expected to deviate in the same way. The calculation performed in the Wilcoxon Signed Ranks Test uses the number of samples analyzed and a ranking of the difference between the result obtained from a technology and the corresponding result from the confirmatory laboratory. The rankings can be compared to predetermined values on a standard Wilcoxon distribution table, which indicates whether, overall, the two methods have produced similar results.

Two of the field screening technologies, those produced by EnSys and by Millipore, produced semi-quantitative results. These technologies produce results that indicate only whether a sample contains PCP above or below a predetermined range. Semiquantitative technologies, by definition, cannot produce Level 3 data. Linear regression analysis and the Wilcoxon Signed Ranks Test cannot be used to compare semiquantitative results. Instead, PRC used a 2 by 2 contingency table and a Fisher's Test. The Fisher's Test determines whether both data sets are correlated. When used in a two-tailed manner, as it was in this case, its formula is usually conservative. Therefore, use of a modified Chi-square formula is recommended (Pearson and Hartley 1976).

The Fisher's Test statistics were compared to the 90 percent significance level obtained from a standard Chi-square distribution table. This comparison indicated whether, overall, there was a correlation between the results of the two methods. If a correlation existed, the technology was considered accurate, capable of producing Level 2 data.

Finally, the precision of each quantitative technology was statistically compared to the precision of the confirmatory laboratory using Dunnett's Test. This test was used to assess whether the precision of the technology and that of the confirmatory laboratory were statistically equivalent. First, the mean RPD for all samples and their respective duplicates analyzed by the confirmatory laboratory was determined. The RPD of each duplicate pair analyzed by each of the technologies

was then statistically compared to this mean. It should be noted that a Dunnett's result showing the precisions are not similar does not mean that the precision of the technology was not acceptable, only that it was different from the precision of the confirmatory laboratory. In particular, Dunnett's Test has no way of determining whether or not any difference between the two data sets actually resulted because a technology's data was more precise than the confirmatory laboratory's. Verification

of the Dunnett's results was provided by the Wilcoxon Signed Ranks Test.

Overall, for this demonstration, the determination of significance for inferential statistics was set at 90 percent. However, regression analysis was considered to show a significant relationship if the  $r^2$  was greater than 0.85 for Level 3 data and between 0.75 and 0.85 for Level 2 data.



## Section 5 Confirmatory Analysis Results

All samples collected during this demonstration were submitted to the EPA Region 7 Laboratory for confirmatory analysis. The water samples were analyzed by the EPA Region 7 Laboratory under the ESAT Contract, and the soil samples were analyzed under the RECAP Contract. The EPA Region 7 Laboratory assigned sample numbers to each sample submitted for analysis. In this manner, analyst bias was eliminated during analysis of the samples. The result for each sample is presented within Sections 6 and 7.

### Confirmatory Laboratory Procedures

EPA Region 7 Laboratory Quality Assurance and Data Evaluation (QADE) Branch personnel conducted a Level II data review on the results provided by the confirmatory laboratory. A Level II data review does not include an evaluation of the raw data or a check of calculated sample values. A review of the raw data and a check of the calculations was performed by QC personnel from the confirmatory laboratory before the data package was submitted to the EPA Region 7 Laboratory QADE Branch. PRC was not able to review the raw data generated. However, PRC did review the laboratory case narratives and the EPA Region 7 Laboratory QADE Branch comments generated by the Level II data review.

The following sections discuss specific procedures used to identify and quantitate semivolatile organic compounds (SVQC), and specifically PCP, using the following methods: SW-846 Method 8270A (soil and water), SW-846 Method 8151A (soil), and EPA Method 515.1 (water).

### Sample Holding Times

All of the analytical methods used for confirmatory analysis require that all sample extractions be completed within 7 days from the time a sample was collected. Due to the stability of PCP, EPA's ORD Methods Validation Section extended these holding time

requirements by 4 days for this demonstration. The analysis of the sample extracts must be completed within 40 days of sample receipt. The holding time requirements for the demonstration's samples were met.

### Sample Extraction

The method used for the extraction of soil samples prior to analysis by EPA Method 8270A was EPA Method 3550. This method involves sonication extraction of the soil using methylene chloride. The confirmatory laboratory used both the low concentration extraction method and the high concentration extraction method discussed in the method. To determine the appropriate extraction method to use for the analysis of individual soil samples, the confirmatory laboratory screened each sample using the screening techniques recommended in EPA Method 8270A. EPA Method 3510A was used for the extraction of water samples and involves a separatory extraction of the water with methylene chloride. To ensure that phenolic compounds, such as PCP, will be adequately extracted from the water samples, two extractions of each water sample were performed. The pH of the water was adjusted to greater than 12 and extracted, then the pH of the water sample was adjusted to below 2 and extracted. The two sample extracts were then combined for sample analysis.

The low-level detection analytical methods for PCP included different procedures for sample extraction. The method used for the soil samples, EPA Method 815 1A, involved an acidification of the soil sample, followed by an ultrasonic extraction with methylene chloride. This extraction is similar to the EPA Method 3550 sonication extraction. The soil sample extract was then taken through an acid-base partition. The acid-base partition was used to remove potentially interfering compounds from the sample extract. The sample extract was then concentrated and taken through a diazomethane derivatization. This procedure replaces the hydrogen atom of the phenolic hydroxide group with a methyl anion. This derivatization removes the polarity associated with

PCP and enables improved chromatographic behavior. PCP standards used for sample identification and quantitation were taken through the same derivatization steps as samples to allow a direct comparison of concentration. That is, no correction factor needs to be used for the molecular weight of the derivatization product.

The low-level detection analytical method used for water samples, EPA Method 515.1, involved a separatory extraction of the water sample with methylene chloride. A pH adjustment of the water samples was performed similar to the pH adjustment used for the water samples extracted with EPA Method 3510A. In EPA Method 515.1, the solvent extract from the basic extraction is discarded because it contains no PCP. This step also removes potential interferences. The water sample extract is then concentrated and derivatized in the same manner as the soil sample extracts. Again, this derivatization removes the polarity associated with PCP and provides improved chromatographic behavior. PCP standards used for sample identification and quantitation were taken through the same derivatization steps as the samples to allow a direct comparison of concentration.

### ***Reporting Limits and Initial and Continuing Calibrations***

The reporting limit for soil samples analyzed by EPA Method 8270A was 0.330 ppm. The reporting limit for soil samples analyzed by EPA Method 8151A was 0.076 ppb. The reporting limit for water samples analyzed by EPA Method 8270A was 50 ppb. The reporting limit for water samples analyzed by EPA Method 515.1 was 0.076 ppb. Method-required initial and continuing calibration procedures were appropriately conducted, and all method-required criteria for these calibrations were met.

### ***Sample Analysis***

The confirmatory laboratory performed sample analysis by first analyzing samples using EPA Method 8270A. Based upon the screening results, the samples were extracted with either the low concentration method or the high concentration method. Samples which did not provide a positive response for PCP with EPA Method 8270A were analyzed by one of two low-level detection methods, EPA Method 8151A for soil samples and EPA Method 515.1 for water samples.

EPA Method 8270A uses gas chromatography for compound separation and a mass spectrometer for identification and quantification of PCP. EPA Method 815 1A uses gas chromatography from compound separation and an electron capture detector (ECD) for

identification and quantification of PCP. The chromatographic column used for EPA Methods 8270A and 815 1A can vary and appropriate columns can be found in the methods.

For EPA Method 8270A, compound identification was required to meet two criteria: (1) the sample component relative retention time was to fall within  $\pm 0.06$  relative retention time units of the standard component, and (2) the mass spectrum of the sample compound was to correspond with the standard compound mass spectrum.

Soil and water samples, which were found to contain no PCP during the EPA Method 8270A analysis, were analyzed using the EPA Method 8151A and 515.1, respectively. PCP identification was made if a sample peak eluted within the retention time window established during the initial calibration.

### ***Quality Control Procedures***

Method blanks are used to monitor the presence of laboratory-induced contamination. The EPA Region 7 Sample Management Office (SMO) provided blank soil and blank water samples for use as method blank samples during the analysis of demonstration samples. An acceptable method blank must not provide a positive response for the target compounds above the reported detection limit. Method blank samples were stored, extracted, and analyzed in exactly the same manner as the demonstration samples. Results for all method blank samples extracted and analyzed along with the demonstration samples were found to be acceptable.

Internal standards were used for the analysis of demonstration samples by EPA Method 8270A. Internal standards were added to all standards, blanks, samples, and QC samples prior to injection into the GC/MS system. The internal standards were used to provide response factors for each of the target compounds. During the analysis of soil samples, seven samples exhibited internal standard responses which were outside of the QC limits of 50 to 150 percent recovery. All of the affected samples provided internal standard responses which were less than 50 percent. The soil samples affected were samples 038, 060, 062, 068, 090, 091, 095. Of these samples, three -- 090, 091, and 095 -- were found to contain no detectable levels of PCP, and no corrective action was taken. Instead, they were reanalyzed using EPA Method 8151A. The remaining samples were reanalyzed to verify that the internal standard response was below 50 percent recovery. The reanalysis showed that internal standard response was below 50 percent recovery. No corrective action was taken by the laboratory, which attributed the low

recovery to matrix effects inherent to the samples. In the Region 7 Laboratory QA/QC Branch review of the data, the same conclusion was reached.

Surrogate standards were used to evaluate the efficiency of the extraction and analysis processes and to evaluate matrix effects. Surrogate standards used for EPA Method 8270A include deuterated standards, which provide a different mass spectrum when compared to the nondeuterated compound. Surrogate standard recoveries for the soil samples all fell within the acceptance ranges. The data review performed by the Region 7 QA/QC Branch indicated that surrogate recoveries for some of the water samples were outside of the acceptance ranges, but no information indicated which samples or how many samples fell outside surrogate recovery acceptance ranges. Corrective action was not taken because the acceptance ranges listed in the method are for advisory purposes only. The surrogate standard used for EPA Method 8151A and EPA Method 515.1 was 2,4-dichlorophenylacetic acid (DCAA). The acceptance range for DCAA was determined by the RECAP and Region 7 Laboratory through a statistical analysis of 30 or more standard surrogate recoveries. The mean and standard deviation were then calculated, and the acceptance range was determined by applying a  $\pm 3$  standard deviations around the mean. All samples analyzed with EPA Method 8151A and EPA Method 515.1 provided surrogate recoveries which fell within the laboratory-generated control limits.

Matrix spike samples were aliquots of original samples into which a known concentration of the target compounds was added. The EPA Region 7 Environmental Services Division specified which samples were to be used as confirmatory laboratory matrix spike samples. The specified soil samples were samples 036,048,053,073, 087, and 098, all analyzed using EPA Method 8270A, and sample 089, analyzed using EPA Method 8151A. The specified water samples were samples 101 and 111, analyzed using EPA Method 515.1, and sample 104, analyzed using EPA Method 8270A. The soil matrix spike samples analyzed using EPA Method 8270A were spiked with all of the target compounds reported by the method. Water sample matrix spike samples analyzed using EPA Method 8270A were spiked with nine of the target compounds reported by the method. Matrix spike samples analyzed with EPA Methods 8151A and 515.1 were only spiked with PCP. Soil matrix spike data for PCP is shown in Table 5-1; water matrix spike data for PCP is shown in Table 5-2. It should be noted that the confirmatory laboratory does not reprepare or reanalyze matrix spike samples.

The soil sample matrix spike recoveries were greatly influenced by the high concentrations of PCP present in the original sample relative to the amount spiked. Only one sample, 098, resulted in recoveries for both the matrix spike and matrix spike duplicate sample which could be considered acceptable. A clear evaluation of the effects of matrix on PCP recovery is not possible due to the high concentrations of PCP in the original sample and the comparatively low levels of PCP added to the matrix spike samples. The water sample matrix spike sample analyzed using EPA Method 8270A resulted in high recoveries. These recoveries are on the high end of the QC acceptance criteria for PCP recoveries listed in EPA Method 8270A (14 to 176 percent recovery). However, the agreement between the matrix spike and matrix spike duplicate was excellent as determined by the RPD of the matrix spike recoveries. The water matrix spike samples analyzed using EPA Method 515.1 were affected by the concentration of PCP in the original sample. Although the matrix spike recoveries for sample 101 were found to be acceptable, the recoveries of PCP spiked into the sample were affected by the much larger concentration of PCP in the original sample. Sample 111 also was affected by the concentration of PCP in the original sample. The results of both the matrix spike sample and the matrix spike duplicate sample were less than the result for the original sample. This may indicate a heterogeneity problem with the sample. The low levels of PCP added to this sample were not enough to obtain an accurate indication of matrix spike recovery.

Blank spike samples were prepared by the EPA Region 7 Laboratory SMO for the water sample analysis performed by EPA Methods 8270A and 515.1. These samples are used to evaluate the accuracy of the laboratory. The blank spike samples were stored, extracted, and analyzed in the same manner as all other samples. The percent recoveries of the blank spike samples fell within the 14 to 176 percent QC acceptance criteria listed in EPA Method 8270A and the 67.6 to 192.4 percent acceptance criteria listed in EPA Method 515.1. The accuracy of the analysis of water samples using EPA Methods 8270A and 515.1 was found to be acceptable, based on the blank spike sample results.

### **Data Reporting**

The data report PRC received from the EPA Region 7 Laboratory included a standard EPA Region 7 Analysis Request Report. Results were reported on a dry-weight basis, as required in the methods. PRC obtained data on the loss-on-drying determination for each of the samples. The loss-on-drying values were used to convert the

TABLE 5-1. SOIL MATRIX SPIKE SAMPLE RESULTS FOR EPA METHODS 8270A AND 8151A

Sample No.	Amount Found in Original Sample (ppm)	Amount Added to Matrix Spike Sample Duplicate (ppm)	Amount Found in Matrix Spike Sample (ppm)	Percent Recovery (%)	Amount Found in Matrix Spike Duplicate Sample (ppm)	Duplicate's Percent Recovery (%)	Relative Percent Difference (%)
036	40.0	1.90	66.0	1,370	51.0	579	81
048	30,000	46.0	22,000	0	24,000	0	0
053	2.30	1.50	12.0	647	5.20	193	108
073	86.0	11.0	130	400	93.0	64	145
087	46.0	1.40	57.0	786	64.0	1,285	48
089	0.247	0.098	0.315	69	0.241	0	200
098	0.70	0.41	0.82	29	0.98	68	80

TABLE 5-2. WATER MATRIX SPIKE SAMPLE RESULTS FOR EPA METHODS 8270A AND 515.1

Sample No.	Amount Found in Original Sample (ppb)	Amount Added to Matrix Spike Sample and Duplicate (ppb)	Amount Found in Matrix Spike Sample (ppb)	Percent Recovery (%)	Amount Found in Matrix Spike Duplicate Sample (ppb)	Duplicate's Percent Recovery (%)	Relative Percent Difference (%)
101	4.14	0.446	4.46	72	4.24	22	106
104	50.0 U	200	348	174	353	177	2
111	1.85	0.398	1.55	0	1.64	0	0

Note:  
 U Not detected above detection limit.

confirmatory laboratory data from a dry-weight basis to a wet-weight basis.

Results were reported by the confirmatory laboratory in micrograms per kilogram ( $\mu\text{g}/\text{kg}$ ) for soil samples and micrograms per liter ( $\mu\text{g}/\text{L}$ ) for water samples. Soil sample results were converted to milligrams per kilogram (microgram/kg) so they could be compared to the results from the technologies, all of which reported results for soil samples in microgram/kg. The results from the technologies for water samples were reported in  $\mu\text{g}/\text{L}$ , so no conversion of the confirmatory laboratory data was needed.

**Data Quality Assessment**

Accuracy refers to the difference between the sample result and the true concentration of compound in the sample. Bias, a measure of the departure from complete accuracy, can be caused by such processes as loss of compound during the extraction process, interferences, and systematic contamination or carryover of a compound from one sample to the next. Accuracy for the confirmatory laboratory was assessed through the use of PE samples. Four of the PE samples used for this demonstration were purchased from Environmental

Research Associates (ERA). Two of these PE samples were soil and two were water. These 'samples contained a known quantity of PCP. ERA supplied data sheets for each PE sample which included the true concentration and an acceptance range for the sample. The acceptance range was based on the 95 percent confidence interval taken from data generated by ERA and EPA interlaboratory studies. A third water PE sample was prepared by the PRC lead chemist to widen the range covered by the PE samples.

The PE samples contained different concentrations of PCP. These samples were extracted and analyzed in the same manner as the other water and soil samples. The confirmatory laboratory did not know which samples were PE samples or the certified values and acceptance ranges. The true value concentration of soil PE sample 099 (the low-level sample) was 7.44 ppm with an acceptance range of 1.1 to 13 ppm. The result reported by the confiitory laboratory for this sample was 4.02 ppm, which was within the acceptance range. The percent recovery of this sample by the confirmatory laboratory was 54 percent. The true concentration of soil PE sample 100 (the high-level sample) was 101 ppm with an acceptance range of 15 to 177 ppm. The result reported for this sample by the confirmatory laboratory

was 52.4 ppm, which was within the acceptance range. The percent recovery of this sample by the confirmatory laboratory was 52 percent.

The true value concentration of water PE sample 106 (the low-level sample) was 68.4 ppb with an acceptance range of 10 to 120 ppb. The result reported by the confirmatory laboratory for this sample was 10.3 ppb, which was within the acceptance range. The percent recovery of this sample by the confirmatory laboratory was 15 percent. The true concentration of water PE sample 107 (the high-level sample) was 2,510 ppb with an acceptance range of 377 to 4,420 ppb. The result reported for this sample by the confirmatory laboratory was 2,050 ppb of PCP, which was within the acceptance range. The percent recovery of this sample by the confirmatory laboratory was 82 percent. The true value concentration of water PE sample 113 (the PE sample prepared by PRC) was 7.50 ppb. No acceptance range was statistically determined for this PE sample. Instead, PRC established a 30 to 170 percent window of acceptable values around the true value result of the low-level PE sample. This window is consistent with both the acceptance ranges of the PE samples prepared by ERA and the QC Acceptance Criteria for PCP recovery stated in EPA Method 8270A. The confirmatory laboratory result for this PE sample was within the acceptance range. Based on the results for all of the PE samples, the accuracy of the confirmatory laboratory was acceptable.

Precision refers to the degree of mutual agreement between individual measurements and provides an estimate of random error. Precision for the confirmatory laboratory results was determined through the use of field duplicate samples. Normally laboratory duplicates are used for this. However, no laboratory duplicates were analyzed by the confirmatory laboratory. Field duplicates are two samples collected together, but delivered to the laboratory with separate sample numbers. Typically, field duplicate samples are used to measure both sampling and analysis error. PRC established control limits for field duplicate RPDs. These control limits are similar to those used to determine matrix spike recovery acceptance control limits. To establish the control limits, all sample pairs that did not produce two positive results were removed from the data set. Then the RPD for each pair was calculated, and the mean RPD and standard deviation were determined. The lower control limit was set at zero because this would mean that the results from a duplicate and its sample matched perfectly. The upper control limit was set by multiplying the standard deviation by two and adding it to the mean RPD. The RPD of each sample pair was then compared to these

control limits. Each sample pair RPD was expected to fall within the control limits.

Fourteen soil field duplicate samples were collected and analyzed by the confirmatory laboratory during this demonstration. Field duplicate samples represented 17 percent of all soil samples collected and analyzed. The original results ranged from 0.10 to 26,100 ppm. The duplicate sample results ranged from 0.09 to 30,260 ppm. RPD values for the soil field duplicate pairs ranged from 1 to 168 RPD. The mean RPD value of the soil field duplicate pairs was 33 percent, with a standard deviation of 47 percent. For the soil field duplicate pairs, the control limits were found to be 0 to 128 RPD. Thirteen of the fourteen, or 93 percent, of the field duplicate sample pairs fell within this range.

Six water field duplicate samples were collected and analyzed by the confirmatory laboratory during this demonstration. Field duplicate samples represented 32 percent of all water samples collected and analyzed. The original results ranged from 0.175 to 1,810 ppb. The field duplicate sample results ranged from 0.63 to 2,020 ppb. RPD values for the water field duplicate pairs ranged from 0 to 113 RPD. The mean RPD value of the water field duplicate pairs was 30 percent with a standard deviation of 41 percent. For the water field duplicate pairs, the control limits were found to be 0 to 112 RPD. Five of the six, or 83 percent of the field duplicate sample pairs, fell within this range.

Overall, this data shows 'excellent agreement between the samples and their respective field duplicates, indicating a high degree of precision by the confirmatory laboratory. The mean RPD also indicated that the method used to homogenize the samples before splitting them for analysis was highly effective.

Completeness refers to the amount of data collected from a measurement process compared to the amount that was expected to be obtained (Stanley and Vemer 1983). The completeness objective for this project was 95 percent. This demonstration resulted in the analysis of 98 soil samples, 14 soil sample duplicates, 2 soil PE samples, 10 water samples, 6 water sample duplicates, and 3 water PE samples. Results were obtained for all of these samples. Completeness for the confirmatory laboratory was 100 percent.

## **Confirmatory Laboratory Costs and Turnaround Times**

The cost for performing PCP analysis by EPA-approved analytical methods varies from laboratory to laboratory. The cost of analysis depends upon the

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number of samples submitted for analysis, the matrix, and the level of QC performed. The following costs are given as general guidelines. EPA Method 8270A analysis costs range from \$250 to \$400 per sample. EPA Method 8151A analysis costs range from \$150 to \$250 per sample. EPA Method 515.1 analysis costs range from \$125 to \$200 per sample. Turnaround times

for samples submitted for analysis with EPA-approved analytical methods range from 14 to 30 days. The turnaround time also depends upon the number of samples submitted for analysis, the matrix, and the level of QC performed. Faster turnaround times may be available for an additional cost.

## Section 6

### EnSys Inc.: Penta RISC Test System

This section provides information on the Penta RISC Test System including background information, operational characteristics, performance factors, a data quality assessment, and a comparison of its results with those of the confirmatory laboratory.

#### Theory of Operation and Background Information

The system is designed to provide quick, semiquantitative results for PCP concentrations in soil and water samples (see Exhibit 6-1). The system is composed of a soil test kit and a water test kit. Soil and water samples must be analyzed separately and compared to different PCP calibrators. The system uses the principles of enzyme-linked immunosorbent assays (ELISA). ELISA-based analytical technologies use antibodies to provide compound-specific reaction, detection, and quantitation. These antibodies are produced by injecting animals, usually rabbits, with either a measured amount of PCP or a PCP analog. The PCP or analog also are known as immunogens. The animal's immune system produces an antibody specific to the PCP or analog in much the same manner that the human immune system produces antibodies to fight infection by common viral and bacterial pathogens. Booster injections of the immunogen are continued until a maximum antibody-binding response is attained. The antibodies are then collected and separated from the animal's blood for use in the manufacture of the immunoassay kit. The developer covalently binds the antibodies to the inside walls of test tubes for use in its test system.

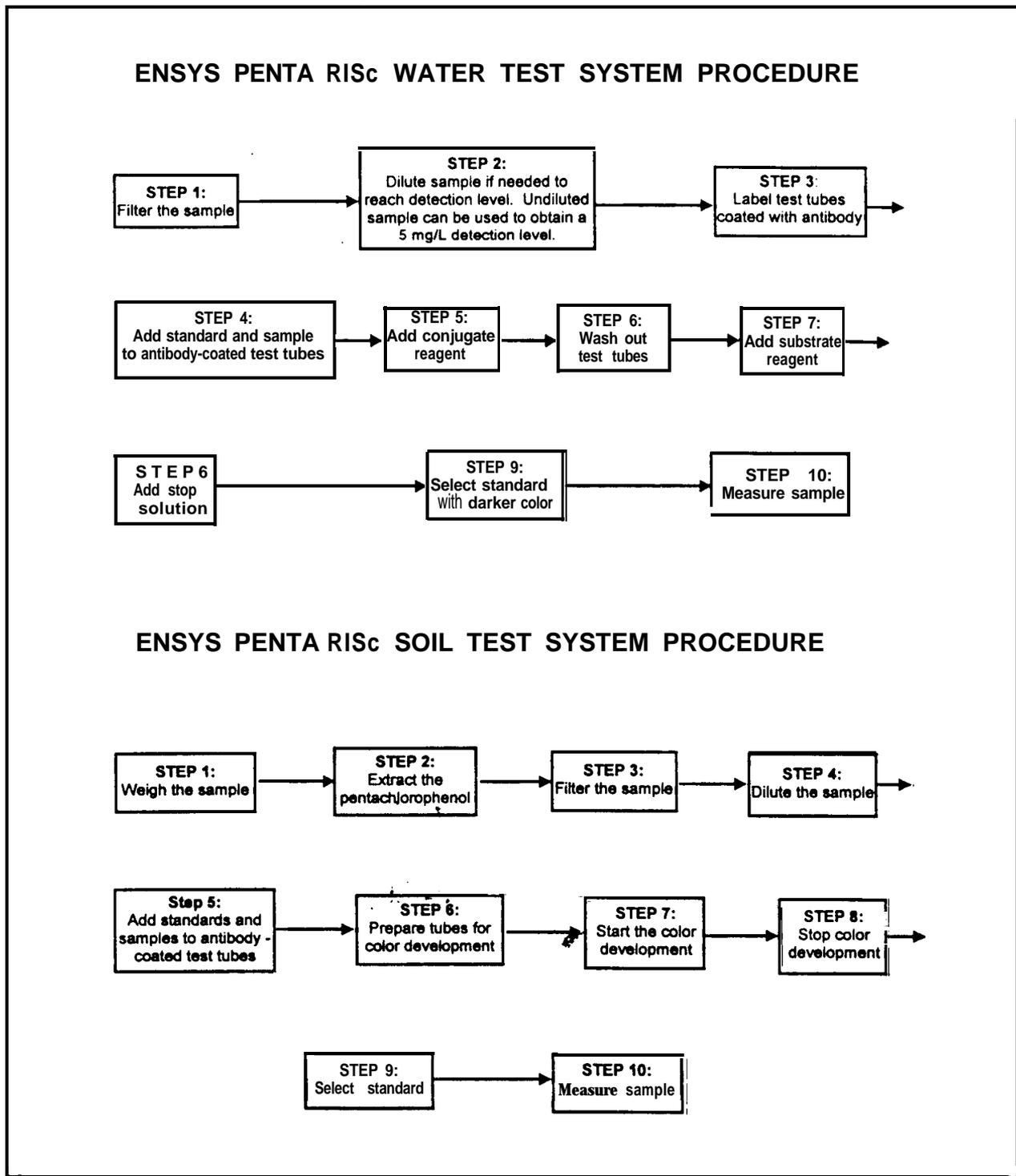
In addition to the antibodies, ELISA-based technologies generally use an enzyme conjugate in the analysis step of the immunoassay test. The enzyme conjugate is formed by covalently binding a PCP analog to a horseradish peroxidase enzyme. This enzyme conjugate competes with PCP in an environmental sample for antibody binding sites on the walls of the test tube. The enzyme conjugate provides the means for

identification and quantitation of PCP. ELISA-based technologies use chromogenic reagents that react specifically with the enzyme conjugate. These results allow identification and quantitation of PCP.

In this technology, a mixture of two substrates reacts with the enzyme conjugate to produce a brilliant blue color. Because an exact number of antibody binding sites are available on each test tube and an exact number of enzyme conjugate molecules are introduced into each sample, the only variable which exists is the number of PCP molecules present in the environmental sample. The PCP in the sample will compete with the enzyme conjugate molecules for antibody binding sites, thus reducing the amount of blue color formed by the reaction of the substrate with the enzyme conjugate molecules. It is the amount of blue color formed by the sample that is used for identification and quantitation of PCP in the environmental sample.

Semiquantitative interpretation is performed by comparing differential photometer readings of the sample to differential photometer readings of a PCP standard taken through the same immunoassay steps required for the samples. If the differential photometer reading of a sample is zero, or a negative value, the sample contains close to or more than the detection level of interest. If the differential photometer reading of a sample is positive value, the sample contains less than the detection level of interest. The use of three sample dilutions allows for a closer approximation of PCP concentrations in samples.

The system is designed to detect PCP, but other compounds may respond to the system. This is referred to as cross reactivity and is measured by determining at what concentration a specific compound will yield a positive result using the system. Another phenomenon that may affect immunoassay tests is masking cross reactivity, which is caused by cross reacting compounds inhibiting or obscuring the reaction of the target compound with the antibodies. The developer has



evaluated a number of compounds and has provided information on cross reactivity. Table 6-1 presents this positive value, the sample contains less than the detection level of interest. The use of three sample dilutions allows for a closer approximation of PCP concentrations in samples.

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compounds may respond to the system. This is referred to as cross reactivity and is measured by determining at what concentration a specific compound will yield a positive result using the system. Another phenomenon that may affect immunoassay tests is masking cross reactivity, which is caused by cross reacting compounds inhibiting or obscuring the reaction of the target compound with the antibodies. The developer has

evaluated a number of compounds and has provided information on cross reactivity. Table 6-1 presents this information. As part of this demonstration, a specificity study was conducted that concerned the test system's reaction to diesel fuel, which is a common carrier for PCP, and to chemicals with molecular structures similar to PCP. Data from the specificity study is presented later in this report.

**TABLE 6-1. CHEMICAL CROSS REACTIVITY AS REPORTED BY ENSYS**

Compound	Concentration Required for a Positive Interpretation by Soil Test System (ppm)	Concentration Required for a Positive Interpretation by Water Test System (ppb)
PCP	0.5	5
Phenol	> 1,000	> 600
4-Chlorophenol	> 1,000	> 600
2,4-Dichlorophenol	> 1,000	> 1,000
2,6-Dichlorophenol	700	600
3,5-Dichlorophenol	NA	> 1,000
2,3,4-Trichlorophenol	400	600
2,4,5-Trichlorophenol	100	500
2,4,6-Trichlorophenol	16	100
Tetrachlorophenol	1.2	7
Tetrachlorohydroquinone	500	> 1,500
Pentachlorobenzene	> 1,000	> 1,400
Aroclor 1254	> 1,000	>100

## Operational Characteristics

Instrumentation required for the system is shipped in a small suitcase-sized, plastic container. This container has a handle and is very easy to carry. The reagents and supplies required for the system are shipped in large plastic boxes. The containers and boxes needed for analysis of 200 samples would fit in the trunk of a large car or in the back seat of a smaller car for transportation to close proximity sites. The containers and boxes can easily be shipped by commercial carriers to sites farther away. During this demonstration, all containers and boxes were shipped to the site directly from the developer.

The system is composed of two test kits: the Penta RISC Soil Test System and the Penta RISC Test System, used for water samples. An accessory kit, containing instrumentation, also is required for the analysis of soil and water samples. The Penta RISC Soil Test System includes the following: (1) four sample extraction jars with screw caps, each with stainless-steel ball bearings and premeasured extraction solvent, (2) four weigh boats, (3) four wooden spatulas, (4) four bulb transfer pipettes, (5) four filtration devices composed of four filtration barrels and four filtration plungers, (6) four

mechanical pipette pistons and four mechanical pipette capillaries, (7) four sample dilution vials labeled 0.5 ppm, (8) four sample dilution vials labeled 5.0 ppm, (9) four sample dilution vials labeled 50.0 ppm, (10) twelve buffer tubes containing premeasured diluent, (11) eight tubes containing standard, (12) twenty antibody-coated test tubes in an aluminized pouch, (13) one bottle of wash buffer solution, (14) one bottle of Substrate A, (15) one bottle of Substrate B, (16) one bottle of stop solution, and (17) one instruction booklet.

The Penta RISC Test System (for water) includes: (1) four filtration devices composed of four filtration barrels and four filtration plungers, (2) four bulb transfer pipettes, (3) one capillary pipette plunger, (4) four capillary pipettes, (5) twelve sample dilution tubes containing premeasured diluent, (6) eight tubes containing standard, (7) twelve antibody-coated test tubes, (8) one bottle of enzyme conjugate solution, (9) one bottle of wash buffer solution, (10) one bottle of Substrate A, (11) one bottle of Substrate B, (12) one bottle of stop solution, and (13) one instruction booklet.

The accessory kit includes the following: (1) a Pocket Pro Series electronic balance, (2) an Artel DP differential photometer, (3) a Model P-250 Poppette Micropipettor, adjustable from 5 to 250 microliters, and (4) a Westbend electronic timer.

Other equipment that is helpful when using the system, which is not supplied by the developer includes protective gloves, laboratory tissue, a permanent marking pen, paper towels, and liquid and solid waste containers.

For this demonstration, the system was operated in a 28-foot trailer located at the former Koppers site. Electricity was supplied to the trailer for the use of air conditioning and to provide lighting. A refrigerator was required to store the enzyme conjugate solution used with the water test kit. The enzyme conjugate supplied with the soil test kit is in a pelletized form bonded within a sucrose tablet and is more stable than the liquid form of the enzyme conjugate. For this reason, the developer states that the soil test kit does not require refrigeration. The developer recommends storage of the soil test kit at room temperature and recommends that these components should not be exposed to temperatures below 0 °C or above 37 °C. The developer recommends storage of the water test kit components at 4 to 8 °C and that reagents should not be stored at or below 0 °C or above 37 °C. Electricity to operate the differential photometer during this demonstration was supplied by the photometer's rechargeable battery. This battery requires 8 to 24 hours of charging and, when fully charged, can perform up to 500 tests without being

recharged nightly by the electricity in the trailer. Electricity required is a 110-volt circuit. A 3-foot by 2-foot hood was used by the operator for performing soil sample extractions. A 4-foot table was used to perform the assay steps, sample dilutions, sample analysis using the differential photometer, and logbook entries and other data documentation.

The operator chosen for analyzing samples using the technology was Mr. Frank Douglas. Mr. Douglas is an employee of PRC. He earned a Bachelor of Science degree in journalism with a minor in science, a Master of Arts degree in English. While at PRC, Mr. Douglas has worked as an editor in PRC's QC system. Mr. Douglas also has worked as a technician on a similar SITE demonstration performed by PRC. In this capacity, he helped explain how immunoassay systems work and helped interpret the statistics that resulted from the test.

Mr. Douglas's training in the use of the test system included a review of the information provided by the developer before the start of the demonstration. Mr. Douglas also received approximately 6 hours of training at the start of the demonstration by Ms. Rhonda Mudd of EnSys. This training included step-by-step procedures for extracting, preparing, and analyzing soil and water samples using the system, and instructions on use of the pipettes and instrumentation. Interpretation of sample results was discussed as well as QC requirements. In addition, Mr. Douglas analyzed soil and water samples using the system under the supervision of Ms. Mudd. Mr. Douglas noted that after he had completed the analysis of both soil and water samples, Ms. Mudd felt confident that he was ready to properly operate the system.

The developer states that this product is intended for use by environmental professionals and requires a minimum of training. Mr. Douglas had no prior experience performing immunoassay testing. Mr. Douglas noted that he found the system very easy to operate. Mr. Douglas noted that the developer has dramatically increased the ease of operation for the system by limiting the amount of pipetting and measuring necessary. He also noted that the developer has premeasured the extraction liquid, dilution liquid, buffer liquid, and standards. The developer also color codes the reagents and numbers the reagents used in the water test kit.

The technology is promoted for use as a portable field system. However, it does require special care and handling in the field to avoid damage. The differential photometer requires care during use, shipping, and

transportation to avoid breaking or damaging internal components. No mechanical or electronic problems were experienced with this equipment during the course of the demonstration.

Instrument reliability for the system was to be evaluated by monitoring the standard calibration responses. QC criteria for the calibration responses included the analysis of a duplicate standard with each standard prepared. The standard which exhibited the higher optical density reading was to be used for sample comparison, and the optical density readings between a standard and its duplicate were not to vary by more than 0.35 optical density units. For the soil system, 46 pairs of standards were prepared. Only three times did the optical density readings between a standard and its duplicate vary by more than 0.35 optical density units. In these cases, all samples analyzed with these pairs were reanalyzed unless their optical density readings were greater than the optical density of the high-level standard. The developer recommended to PRC that samples which gave optical density readings that were greater than the high-level standard did not have to be reanalyzed because, even when reanalyzed with another acceptable calibration, these samples would still give the same result. For the water system, 37 standard pairs were prepared. None of these pairs produced optical density readings which varied by more than 0.35 optical density units.

Two times during the analysis of soil samples the operator of the system noted that a sample turned bright orange upon the addition of the stop solution and produced an extremely high optical density reading. The operator contacted the developer with this problem. EnSys personnel commented that they had seen this problem before and that the sample needed to be reanalyzed. Ms. Mudd referred to this phenomena as a "high flyer" and speculated that it may be attributed to a higher than normal amount of enzyme conjugate in the pellet. These two samples were reanalyzed. The operator of the system also noted that two samples provided inconsistent results. In both cases, the technology reported results less than 5 mg/kg but greater than 50 mg/kg. These samples were reanalyzed, and acceptable results were obtained from the reanalysis.

Immunoassays are enzymatic reactions and can be effected by changes in ambient temperatures. The technology seemed to be affected by the temperature in the trailer during this demonstration. The operator of the system noted several times during the course of the demonstration that analysis problems occurred most frequently in the afternoon. Although PRC did not record temperatures in the trailer during the demonstration, the technology operators noticed a

temperature increase in the trailer during the afternoon. One operator noted that he believed the temperature in the trailer increased 5 to 10 °F in the afternoon. This was noticed even while the trailer was air conditioned. Although this problem seemed to affect standards and QC criteria, the problems associated with the temperature increase were found to have an insignificant effect on the results obtained from the system. However, PRC believes that use of the test system is improved when used in a temperature-controlled environment. This will help minimize problems associated with extremes of temperature or humidity which may affect the immunoassay chemistry.

The test system contains various chemicals in quantities ranging from parts per million levels to 500 milliliters. The more dangerous chemicals include methanol, a flammable solvent and poison, and sulfuric acid, a strong acid. Other chemicals include parts per million levels of PCP. Gloves, safety glasses, and protective laboratory coats are recommended when using the system and were used during this demonstration.

Both the soil and water test kits contain enough reagents and supplies to analyze four samples and can be purchased from the developer for \$225 each. The developer states that either of these test kits can be modified to provide the user with the specific detection level of interest for no additional charge. The developer offers accessory items required for the soil and water systems. The differential photometer is required for both systems and is available for \$935. The balance is available for \$100, and the mechanical pipet is available for \$219. All of these items are required for the soil system. A timer also is sold by EnSys for \$29.95. EnSys offers an accessory pack that includes the differential photometer, the mechanical pipette, the balance, and the timer. This accessory pack costs \$1,250. The accessory pack can be rented from EnSys on a daily or weekly rate. The daily rental rate is \$150 and the weekly rate is \$400. According to EnSys, the shelf-life of the reagents used for its technology is 4 months for the water test kit and 3 months for the soil test kit. An expiration date and lot number is printed on each batch of reagents produced.

Logistical costs will vary depending on the scope of the project. As discussed earlier, the system can be operated outdoors or indoors, with electricity or without. A refrigerator or cooler is required for storage of reagents used with the water system. The best results will be attained when the system is used indoors in a temperature-controlled environment. Logistical costs may include trailer rental, electrical hookup fees, refrigerator rental or purchase, and electrical or gas usage. Waste disposal is another operating cost of the

system. During this demonstration, about 200 samples were analyzed using the test system. The waste generated by these analyses filled half a 55-gallon drum. The appropriate way to dispose of this waste is through an approved incinerator facility. The cost for disposal of one drum of this waste is estimated at \$1,000.

## **Performance Factors**

The following paragraphs describe performance factors, including detection limits, sample throughput, linear range, and drift. Specificity, another performance factor, is discussed separately because of its complexity.

### ***Detection Limits***

The detection limit of the system for water samples is 0.005 ppm. This is based on the analysis of 200 microliters of a water sample and 5 drops of an unspecified amount and concentration of a PCP standard. If the water sample's optical density is greater than that of the PCP standard, then the sample contains less than 0.005 ppm. The detection limit of the system for water samples is greater than the 1.0 ppb maximum contaminant level (MCL) for PCP.

The detection limit of the system for soil samples is 0.5 ppm. This is based on the analysis of 100 microliters of a soil extract (10 grams to 10 milliliters) and an unspecified amount and concentration of a PCP standard. If the soil sample's optical density is greater than that of the PCP standard, then the sample contains less than 0.5 ppm.

### ***Sample Throughput***

Sample throughput was determined by evaluating both the time required to extract and analyze one sample and the number of samples analyzed in 1 work day. According to the developer, about 20 minutes is required to analyze a water or soil sample. When run in batches of four samples, the developer states that analysis of all four samples can be completed in less than 30 minutes. EnSys estimates that 50 to 75 samples can be processed in 1 day. The analysis of samples during this demonstration was completed in 8 days. The average work day was 10 hours in duration. The number of samples analyzed during the demonstration was 184. This number included field duplicate samples, specificity samples, and QC samples. The 184 samples were analyzed in 80 hours or slightly more than two samples per hour. The operator was able to average a sample every 27 minutes, slightly longer than the 20 minutes claimed by the developer. The average number of samples analyzed in a 10-hour work day was 23. This was less than the 50 to 75 samples the developer

estimates can be processed per day. The largest number of samples analyzed in one 10-hour day was 40. The operator noted that the maximum number of soil samples that could be extracted and analyzed in one 10-hour day was 40. This equals one sample every 15 minutes, which is less than the 20 minutes claimed by the developer. The operator also noted that 33 water samples were completed in one 10-hour work day, and that three dilutions of the water samples were prepared, rather than the one dilution usually required. When analyzing water samples with one dilution of the water sample, a much higher sample throughput can be expected. The operator noted that sample analysis time did not include the time required for sample handling, data documentation, difficult extractions, or the preparation of QC samples. The time required by the operator to perform these tasks prevented him from completing analysis of 50 to 75 samples per day, as estimated by the developer.

### **Linear Range and Drift**

This technology uses one level of standard for both water and soil analysis. There is no linear range established for the standards. Samples are serially diluted, 1 to 10, to obtain different levels of detection. Drift normally is a measurement of an instrument's variability in quantitating a known amount of a standard. This technology eliminates the variability associated with drift by requiring that a new standard be analyzed with each set of samples analyzed. A common method for evaluating drift for immunoassays is to compare the responses of standards to a negative control standard. A negative control standard is not required for sample analysis when using the system, and this method of drift evaluation could not be performed during this demonstration.

### **Specificity Study**

Specificity refers to a technology's ability to identify and quantitate a particular contaminant when in the presence of other chemicals that could act as interferants. For this technology, interferants might be natural chemical present in a matrix, carriers or other chemicals used to introduce PCP during wood treatment processes, or other contaminants that might be present at such facilities. To assess the specificity of this technology, PRC studied its chemistry, reviewed its developer's literature, and conducted a specificity study.

Organic sample matrix effects for this technology are primarily caused by humic acids. Humic acids exist in most soils and are found in highest concentrations in topsoil. Because they are slightly polar, humic acids can leech from soil and, therefore, are found in water

samples as well. High concentrations of humic acids in a sample can inhibit the extraction of PCP through absorption. Inorganic sample matrix effects are primarily caused by pH, salinity, and inorganic chemical composition. The pH of water samples may affect the immunoassay chemistry of the system. The pH of water samples should be measured and, if extremes are noted, the sample may need to be neutralize. Salinity of water may sometimes affect immunoassay results. The developer provides no information about how salinity may affect its system. Some inorganic chemicals may have an effect on the system's performance by inhibiting the binding of PCP or the enzyme conjugate to the antibody binding sites. The developer furnished no information concerning the effects of inorganic chemicals.

The operation at the former Koppers site used an isopropyl ether butane carrier for PCP application. The operation at the Winona Post site used a diesel fuel-based carrier for PCP application. One objective of this demonstration was to evaluate the technology's ability to quantitate PCP concentrations in samples contaminated with these carriers. Both solvents are highly volatile and will not have long residence times in surficial soils. The developer reports that diesel fuel in concentrations as high as 10 percent will have no effect on soil results and that concentrations as high as 10 ppm will have no effect on water results. In addition to these PCP carriers, other wood-preserving agents are used in conjunction with PCP. The developer has determined that its system is not affected by concentrations of 1,000 ppm creosote in soil or 1,000 ppb creosote in water, nor by concentrations of 1,000 ppm CCA in soil or 10,000 ppb CCA in water. Another site-specific matrix effect is the presence of other chemicals in the samples. Historical data revealed the presence of other phenols, including chlorophenols, at the sites. It also revealed the presence of dioxins and furans, particularly the octa-isomers, at the sites, but the effects of these chemicals on the technology are believed to be insignificant compared to those of the chlorophenols.

Immunoassays are sometimes depicted as a lock-and-key type of chemical interaction. The lock system is the antibody, and it is designed to interact with the key, PCP or the enzyme conjugate. However, the antibody also will interact with other chemicals. This is referred to as cross reactivity and can be evaluated by determining the concentration of a specific chemical which will provide a positive result when analyzed using the technology. The developer has experimentally determined the cross reactivity for a number of compounds which have similar chemical properties to PCP. This cross reactivity data was presented earlier on Table 6-1. Although the table shows that the system is

very specific to PCP, chemicals other than PCP can provide a positive result. Chlorinated phenols, especially the tetrachlorophenols and the trichlorophenols, have the highest cross reactivities. The system will provide false positive results in samples containing other chlorophenols in high concentrations. Many other industrial and natural chemicals have not been evaluated to determine cross reactivity, and for this reason approved methodologies should be used to confirm positive results.

The specificity study was conducted to determine if this technology would show cross reactivity to several chemicals other than PCP. The specificity study involved spiking clean sand with known concentrations of chlorophenols and diesel fuel. It also involved spiking blank water with a mixture of PCP in diesel fuel. The specificity study's samples were prepared by the lead chemist and distributed to the technology operator along with the demonstration samples. The operator knew that the samples were for the specificity study but did not know what chemical each sample was spiked with, nor the concentration.

The soil specificity samples were prepared by weighing 10 grams of clean sand into a soil extraction bottle and spiking it with microliter amounts of chemical standards. One soil specificity sample, SS-05, was an unspiked sand sample to ensure that the sand would not provide a positive response. Soil specificity samples SS-14 through SS-17 were spiked with 100 ppm of diesel fuel, and soil specificity samples SS-06 through SS-09 were spiked with 10 ppm of 2,4-dichlorophenol. None of these samples produced a positive result.

Soil specificity samples SS-01 through SS-04 were spiked with 10 ppm of 2,3,4,6-tetrachlorophenol. These samples produced greater than 5 ppm, but less than 50 ppm results. The developer states that 1.2 ppm of 2,3,4,6-tetrachlorophenol in a soil sample is required to produce a positive result, and the specificity study supports this claim. Soil specificity samples SS-10 through SS-13 were spiked with 10 ppm of 2,4,6-trichlorophenol. These samples all provided a greater than 0.5 ppm, but less than 5 ppm result. The developer states that 16 ppm of 2,4,6-trichlorophenol in a soil sample is required to produce a positive result with the system, but the study showed that the system will respond to concentrations of 10 ppm of 2,4,6-trichlorophenol.

The water specificity samples were SS-18 through SS-21 and were spiked with 125 ppm of diesel fuel and 50 ppb of PCP. This was done to evaluate the effects of diesel fuel on the recovery of PCP in water samples. The results for these samples were the expected results,

greater than 5 ppb, but less than 500 ppb.

## **Intramethod Assessment**

Intramethod measures of the technology's performance included its results on reagent blanks, the completeness of its results, its intramethod accuracy, and its intramethod precision. Reagent blank samples were prepared by taking reagents through all extraction, cleanup, and reaction steps of the analysis. An acceptable reagent blank sample must not provide a positive result for PCP. Six soil and one water reagent blank samples were analyzed during the demonstration, and none of these reagent blank samples provided a positive response. For this demonstration, completeness refers to the proportion of valid, acceptable data generated. Results were obtained for all of the samples; therefore, completeness was 100 percent.

Intramethod accuracy was assessed by using PE samples and matrix spike samples. Five PE samples were analyzed during the demonstration, two for the soil matrix, and three for the water matrix. Both of the soil samples and two of the water samples were purchased from ERA; the other water sample was produced by PRC. These samples were extracted and analyzed in the same way as all other samples. The operator did not know the samples were PE samples, nor did the operator know the true concentrations or the acceptance ranges.

The true value concentration of soil PE sample 099 was 7.44 ppm with an acceptance range of 1.1 to 13 ppm. The result reported by the technology for this sample was greater than 5 ppm, but less than 50 ppm. The true value concentration of soil PE sample 100 was 101 ppm, with an acceptance range of 15 to 177 ppm. The system indicated the concentration was greater than 50 ppm. These results agree with the true values. The true value concentration of water PE sample 106 was 68.4 ppb, with an acceptance range of 10 to 120 ppb. The technology indicated the concentration was greater than 5 ppb, but less than 500 ppb. The true value concentration of water PE sample 113, the PE sample prepared by PRC, was 7.50 ppb, with an acceptance range of 2.25 to 12.8 ppb. The technology indicated that the concentration was greater than 5 ppb, but less than 500 ppb. In both of these cases, the system's results agreed with the true results. However, the true value concentration of water PE sample 107 was 2,510 ppb, with an acceptance range of 377 to 4,420 ppb. The technology indicated that the concentration was greater than 5,000 ppb, which was above the upper acceptance limit. The accuracy of the PE sample results, therefore, was 100 percent for the soil results and 67 percent for the water results. Overall, the accuracy as measured by the PE samples was 80 percent. The technology could

only have been more accurate if all results fell within the PE sample ranges. Still, the demonstration's criteria stated that more than 90 percent of the samples had to have acceptable results, and the technology, therefore, was not considered accurate. Based on this data the performance of the technology on analyzing PE samples was unacceptable.

Matrix spike and matrix spike duplicate samples also were used to assess intramethod accuracy. Matrix spike samples are aliquots of an original sample into which a known concentration of PCP is added. Original samples to which the spike solution was added were required to contain much less PCP than the amount added. These samples were then extracted and analyzed. Each also was duplicated. Six soil samples and one water sample were used for matrix spike samples. Therefore, there were 12 matrix spike results for soil samples and two for water samples. Soil samples were spiked with 2.00 ppm of PCP. An accurate result for a soil matrix spike sample would be greater than 0.5 ppm, but less than 5 ppm. Of the 12 soil matrix spike samples, 11 provided accurate results. The matrix spike duplicate of sample 003 was found to contain greater than 5 ppm, but less than 50 ppm, overestimating the amount of the PCP spiked into the sample. Overall, the accuracy of the results for the soil matrix spike samples was found to be 92 percent, and the system's performance on the matrix spike analysis was assessed as acceptable. Water matrix spike samples were spiked with 50 ppb of PCP. An accurate result for a water matrix spike sample would be greater than 5 ppb, but less than 500 ppb. Both of the water matrix spike samples provided results of greater than 5 ppb, but less than 500 ppb. The accuracy of the water matrix spike samples, therefore, was found to be 100 percent.

Precision is evaluated by determining the number of duplicate sample results which agreed with the original sample results. Precision was assessed by comparing the results obtained on duplicate samples. Three types of precision data were generated: data from laboratory duplicate samples, data from field duplicate samples, and data from matrix spike duplicate samples. Usually these duplicate samples are used to determine matrix variability and the effects of using several operators. To use the duplicates to measure the method's precision, PRC both controlled for matrix variability by thoroughly homogenizing the samples and controlled for operator effects by using only one operator

Laboratory duplicate samples are two analyses performed on a single sample submitted for analysis. Laboratory duplicate samples were analyzed with each set of 20 samples submitted for analysis. Laboratory duplicate samples were analyzed after the original

sample results were obtained. Only original samples with positive results were used for laboratory duplicate analysis. Six soil and one water laboratory duplicate samples were performed during the demonstration. Of the six soil laboratory duplicate samples analyzed, five were found to agree with the original results. Sample 045 had a result of greater than 5 ppm, but less than 50 ppm the first time it was analyzed and a result of greater than 50 the second time it was analyzed. Only one water laboratory duplicate sample was analyzed during the demonstration. The result for both the original sample and its duplicate were found to be greater than 5 ppb, but less than 500 ppb.

Field duplicates are two samples collected together, but delivered to the laboratory with separate sample numbers. Fourteen soil field duplicate samples were collected and analyzed during this demonstration. Field duplicate samples represented 17 percent of all soil samples collected and analyzed. Of the 14 soil field duplicate samples analyzed, 11 were found to agree with the original results, and 3 were found not to agree with the original results. Ten water field duplicate samples were collected and analyzed during the demonstration. Field duplicate samples represent 43 percent of all water samples collected and analyzed during the demonstration. All 10 water field duplicates were found to agree with the original results.

For this demonstration, precision was considered acceptable if 90 percent of the duplicate pairs provided the same result. Overall, 20 soil duplicate pairs were analyzed during this demonstration, and 16 pairs provided matching results. The precision of the technology during this demonstration was found to be 80 percent. This is below the 90 percent criteria established as acceptable precision. Eleven water duplicate pairs were analyzed during this demonstration, and all 11 provided matching results. The precision of the water system during this demonstration was found to be 100 percent.

Six soil matrix spike duplicate samples and one water matrix spike duplicate sample were analyzed and their results were compared to the results of their respective matrix spike samples. Precision was evaluated by determining the number of matrix spike duplicate pairs which provided the same result. Five of the six matrix spike duplicate results agreed with their respective matrix spike samples. The precision of the soil matrix spike samples was determined to be 83 percent, below the 90 percent criteria. It should be noted that, for the soil matrix spike duplicates to meet the precision criteria, all six of the duplicate results would have had to agree with the matrix spike results. Only one water matrix spike duplicate pair was analyzed

during this demonstration. The matrix spike and matrix spike duplicate results were found to agree, and the precision of the water system, therefore, was found to be acceptable.

### **Comparison of Results to Confirmatory Laboratory Results**

The following paragraphs detail the accuracy and precision of the data from analyses using the Penta RISC Test System when compared to that of the confirmatory laboratory. The results from the confirmatory laboratory are considered accurate, and its precision is considered acceptable. The results for soil and water sample analysis are summarized in Tables 6-2 and 6-3. The results from the soil and water sample analyses will be discussed separately. Within each of these two sample matrices the data will be examined as a whole and according to the site from which the samples were collected.

### **Accuracy**

To assess the accuracy of this technology, PRC compared its data to the data from the confirmatory laboratory by using a 2' by 2' contingency table and a Fisher's Test, as approximated by a corrected Chi-square statistic. This statistic was used to test the null hypothesis that the frequency of correct assays is independent of the analytical method used to produce them. In other words, this statistic can be used to determine whether two test methods produce statistically similar results. This test loses most of its statistical power if any expected frequencies fall below 5. A 2' by 2' contingency table was set up for the overall data set, the site-specific data sets, and for each range evaluated by the technology. The contingency tables were set up to compare the number of correct and incorrect results from a test kit relative to the confirmatory laboratory's data. A Fisher's Test, at a 90 percent confidence level, was then used to determine whether a relationship existed between the two sets of results. Accuracy in this section is defined as relating to the number of correct results. While false positives may not impact the intended application of this technology, they are not correct results.

### **Soil Data Set**

This data set consisted of 114 matched pairs of data. It included two PE samples, 67 samples from the former Koppers site and 45 samples from the Winona Post site. The Penta RISC Test System places analytical data into four categories: (1) below 0.5 ppm, (2) between 0.5 and 5 ppm, (3) between 5 and 50 ppm, and (4) greater than 50 ppm.

Examination of the entire data set revealed that the soil test kit gave 83 correct results and 31 incorrect results. The Fisher's Test showed that results from the two analytical methods were statistically different. The lack of correlation indicates that for the entire data set the technology's results were not accurate. These findings did not change when the entire data set was divided by where the samples had been collected. The technology produced 45 correct results and 22 incorrect results on samples from the former Koppers site, and 37 correct results and 8 incorrect results on those from the Winona Post site.

The confirmatory laboratory found that 14 samples had PCP concentrations of less than 0.5 ppm. For this range, the technology produced 12 samples in this range that had concentrations above 0.5 ppm. The Fisher's Test showed that results from the two sets of data were not statistically similar. The lack of correlation indicates that within this range, the technology's results were not accurate. These findings did not change when the entire data set was divided by where the samples were from. The confirmatory laboratory placed nine samples from the former Koppers site in this range; the technology placed 13 there, nine of them correct and four incorrect. The confirmatory laboratory placed three samples from the Winona Post site in this range; the technology placed eight there, three correct and five incorrect. Due to the small data sets in this range for both the Koppers and Winona Post site samples, this statistical test has little power and at best may indicate a trend of greater accuracy for detecting PCP in an isopropyl ether and butane carrier.

The confirmatory laboratory placed 30 samples into the range between 0.5 and 5 ppm. The technology produced 12 correct results and 23 incorrect results for the entire data set. Eighteen times it placed samples that should have been in this range into a different range and five times it placed samples into this range that should have been elsewhere. The Fisher's Test showed correct results. Two times it incorrectly placed samples that should have been in this range into a different range, and it incorrectly placed seven that results from the two sets of data were not statistically similar. The lack of correlation indicates that within this range, for all data, the technology's results were not accurate. These findings did not change when the entire data set was divided into former Koppers site data and Winona Post site data. The technology produced 11 correct results and 15 incorrect results for the former Koppers site data. The confirmatory laboratory had 24 results in this range. The confirmatory laboratory placed six samples from the Winona Post site into this range; the technology produced one correct result and seven incorrect results.

TABLE 6-2. SEMIQUANTITATIVE PENTA RISc TEST DATA AND CONFIRMATORY DATA FOR SOILS'

Sample No.	Penta RISc Test System (ppm)	Confirmatory Laboratory (ppm)	Technology Accuracy	Sample No.	Penta RISc Test System (ppm)	Confirmatory Laboratory (ppm)	Technology Accuracy
001	>0.5<5	4.20	Correct	028	<0.5	0.45	Correct
001D	>5<50	4.18	FP	029	>0.5<5	1.06	Correct
002	<0.5	1.64	FN	030	>50	28.60	FP
003	<0.5	0.13	Correct	030D	>50	29.00	FP
004	>5<50	2.04	FP	031	<0.5	1.43	FN
005	>50	3.70	FP	032	<0.5	0.62	FN
006	>5<50	1.89	FP	033	<0.5	0.40	Correct
007	>5<50	2.66	FP	034	>0.5<5	0.31	FP
008	>0.5<5	0.66	Correct	035	>50	145.0	Correct
009	>0.5<5	3.52	Correct	036	>50	36.80	FP
010	>50	435.0	Correct	037	>0.5<5	1.19	Correct
011	>50	106.0	Correct	038	>50	77.00	Correct
011D	>50	112.0	Correct	039	>0.5<5	3.32	Correct
012	<0.5	0.056	Correct	040	>50	400.0	Correct
013	>50	32.80	FP	040D	>50	34.40	FP
014	>50	99.60	Correct	041	>5<50	6.44	Correct
015	>50	1,190	Correct	042	>5<50	4.09	FP
016	>50	273.0	Correct	043	>50	655.0	Correct
017	>50	1,335	Correct	044	>50	6,956	Correct
018	>5<50	2.13	FP	045	>5<50	22.10	Correct
019	>5<50	6.89	Correct	046	>0.5<5	0.95	Correct
020	<0.5	0.10	Correct	047	>50	13,920	Correct
020D	<0.5	0.09	Correct	048	>50	26,100	Correct
021	>50	5,320	Correct	048D	>50	30,260	Correct
022	>0.5<5	1.85	Correct	049	>50	255.0	Correct
023	>5<50	1.86	FP	050	>5<50	2.16	FP
024	>0.5<5	1.57	Correct	050D	>0.5<5	1.25	Correct
025	>50	593.0	Correct	051	<0.5	0.43	Correct
026	<0.5	0.42	Correct	052	>50	28.20	FP
027	>5<50	11.30	Correct	053	>0.5<5	2.23	Correct

Sample No.	Penta RISC Test System (ppm)	Confirmatory Laboratory (ppm)	Technology Accuracy	Sample No.	Penta RISC Test System (ppm)	Confirmatory Laboratory (ppm)	Technology Accuracy
054	<0.5	0.47	Correct	076	>50	4,590	Correct
055	>50	3,135	Correct	077	>50	2,040	Correct
055D	>50	3,003	Correct	078	>50	1,720	Correct
056	>50	9.90	FP	079	>50	792.0	Correct
057	>5<50	8.74	Correct	080	>50	2,550	Correct
058	>5<50	3.53	FP	081	>50	125.0	Correct
058D	>0.5<5	9.13	FN	082	>50	2,400	Correct
059	>50	9,600	Correct	083	>50	270.0	Correct
059D	>50	102.6	Correct	084	>50	1,140	Correct
060	>50	1,008	Correct	085	>50	57.70	Correct
061	>50	2,744	Correct	086	>0.5<5	6.59	FN
062	>50	138.0	Correct	086D	>0.5<5	6.88	FN
063	>50	1,610	Correct	087	>50	34.00	FP
064	>50	1,978	Correct	087D	>50	51.80	Correct
065	>50	1,577	Correct	088	>0.5<5	2.58	Correct
066	>50	57.80	Correct	089	>0.5<5	0.21	FP
067	>50	110.0	Correct	090	<0.5	0.55	FN
068	>5<50	47.70	Correct	091	<0.5	0.28	Correct
069	>50	798.0	Correct	092	<0.5	0.57	FN
070	>50	2,888	Correct	093	<0.5	0.19	Correct
071	>50	289.0	Correct	094	<0.5	1.02	FN
072	>50	336.0	Correct	095	<0.5	0.088	Correct
073	>50	74.80	Correct	096	>50	59.80	Correct
073D	>50	78.20	Correct	097	>5<50	14.60	Correct
074	>50	836.0	Correct	098	<0.5	0.57	FN
074D	>50	1,520	Correct	099	>5<50	4.02	FP
075	>50	3,692	Correct	100	>50	52.40	Correct

Notes:

a Samples 1 through 58D were collected from the former Koppers site; Samples 59 through 98 were collected from the Winona Post site. Samples 99 and 100 were PE samples.

FP False positive.

FN False negative.

J Reported amount is below detection limit or not valid by approved QC procedures.

ND PCP was not detected above the detection limit.

NA Either the technology, the confirmatory laboratory, or both, did not detect PCP.

TABLE 6-3. SEMIQUANTITATIVE PENTA RISc DATA AND CONFIRMATORY DATA FOR WATER'

Sample No.	PENTA RISc Test System (ppm)	Confirmatory Laboratory (ppm)	Technology Accuracy
101	>0.005<0.5	0.004	False Positive
102	>5	15.900	Correct
103	>5	13.500	Correct
104	>5	0.012	False Positive
105	>0.005<0.5	0.849	False Negative
105D	>0.005<0.5	0.640	False Negative
106	>0.005<0.5	0.010	Correct
107	>5	2.050	False Negative
108	>0.005<0.5	0.002	False Positive
108D	>0.005<0.5	0.002	False Positive
109	>0.005	0.000	False Positive
109D	>0.005	0.001	False Positive
110	>0.005<0.5	0.018	Correct
110D	>0.005<0.5	0.018	Correct
111	>0.005	<0.001	False Positive
111D	>0.005	<0.001	False Positive
112	>5	1.810	False Positive
112D	>5	2.020	False Positive
113	>0.005<0.5	0.002	False Positive

## Note:

Samples 101 through 105D were collected from the Winona Post site; samples 108 through 112D were collected from the former Koppers site. Samples 106, 107 and 113 were PE samples.

Due to the small data set in this range for the Winona Post site samples, this statistical test has little power and at best may indicate a trend of greater accuracy for PCP in an isopropyl ether and butane carrier.

The confirmatory laboratory placed 18 samples into the range between 5 and 50 ppm. The technology produced seven correct results and 21 incorrect results. Eleven times it placed samples that should have been in this range into other ranges, and 10 times it placed samples into this range that should have been elsewhere. The Fisher's Test showed that results from the two sets of data were not statistically similar. The lack of correlation indicates that within this range, for all data, the technology's results were not accurate. These findings did not change when the entire data set was divided into the former Koppers site data and the Winona Post site data. The technology produced five correct results, and 16 incorrect results for the former

Koppers site data. When compared to the 13 results the confirmatory laboratory had in this range, the former Koppers site results were statistically different from the confirmatory laboratory's data. The technology produced two correct results and three incorrect results on samples from the Winona Post site. The confirmatory laboratory had five results in this range. Due to the small data set in the range for the Winona Post site samples, this statistical test has little power and at best may indicate a trend of no carrier effect.

The confirmatory laboratory reported that 52 samples had concentrations of PCP greater than 50 ppm. The technology produced 52 correct results and nine incorrect results. All of the incorrect results were due to samples incorrectly being placed into this range. The Fisher's Test showed that results from the two sets of data were not statistically similar. The lack of correlation indicates that within this range, for all data,

the technology's results were not accurate. These findings changed when the entire data set was divided into the former Koppers site data and the Winona Post site data. The confirmatory laboratory placed 20 samples from the former Koppers site into this range. The technology produced 20 correct results and 8 incorrect results for this site. The Fisher's test showed that the two sets of data from the former Koppers site were statistically different. Therefore, for the former Koppers site samples, the technology's results were not accurate. The confirmatory laboratory placed 31 samples collected from the Winona Post site into this range. The technology produced 31 correct results and one incorrect result. When these results were compared, no statistically significant difference was seen between the two data sets. Therefore, for the Winona Post site samples in this range, the results were accurate.

Overall, 83 of 114 times the technology was correct. This is 73 percent of the time. Of the other 31 times, the technology gave 21 false positive results and nine false negative results. This equates to an 18 percent false positive rate and a 9 percent false negative rate. All of the false negative results were produced when samples containing less than 10 ppm of PCP were analyzed.

When the former Koppers site data is examined alone relative to the confirmatory laboratory's results, 45 of 67 times the technology was correct. This is 68 percent of the time. Of the other 22 times, the technology gave 18 false positive results and four false negative results. This equates to a 26 percent false positive rate and a 6 percent false negative rate. When the Winona Post site data was examined alone relative to the confirmatory laboratory's results, 37 of 45 times the technology was correct. This is 82 percent of the time. Of the other eight times, the technology gave two false positive results and six false negative results. This equates to a 4 percent false positive rate and a 13 percent false negative rate.

Overall, for the soil matrix the technology is conservative. It was not always accurate, relative to the confirmatory laboratory. The semiquantitative nature of the technology does not allow it to be placed in a Level 3 data quality category. Based on the developer's QA requirements and performance specifications for this technology's use, it can produce Level 2 data. However, the technology never met its developer's specifications for percentages of correct, false positive, and false negative results, and it exhibited a lack of correlation to the confirmatory laboratory. The developer's performance criteria are: (1) less than 12 percent false positives, (2) less than 1 percent false negative results, and (3) greater than 88 percent correct

results. The failure to meet its developer's specifications and its lack of correlation to the confirmatory data places this technology in a Level 1 data quality category. Because the false negatives all occurred in samples containing less than 10 ppm PCP, this technology may be applied to sample characterization for applications that require accuracy at concentrations greater than 10 ppm PCP. In these cases, false negatives would most likely occur below target levels and other errors (false positives) would result in a conservative interpretation of data. Therefore, the technology could be used to assist in some activities.

The technology had a slightly higher percentage of correct readings when used on the samples from the Winona Post site. The former Koppers site samples produced a higher percentage of false positives, while the Winona Post samples provided a higher percentage of false negatives.

## Water Data Set

This data set consisted of 19 matched pairs of data. This data set included three PE samples, five duplicate sample pairs from the former Koppers site, and five samples and one duplicate sample pair from the Winona Post site. Due to the small data set size, PRC considered duplicate pairs as two individual samples for this assessment. Therefore, the data set as a whole consisted of 19 data points while the data set for the former **Koppers** site samples contained only 10 samples, and the data set for the Winona Post site samples contained only six samples. The data sets for the individual sites, when divided into the technology's analysis ranges, are too small to give any quantitative statistical analysis meaningful power. Because of this, PRC only evaluated the entire data set. Significance within the individual concentration ranges was not evaluated though trends will be identified.

The results from this technology place analytical data into four categories: (1) below 0.005 ppm, (2) between 0.005 and 0.5 ppm, (3) between 0.5 and 5 ppm, and (4) greater than 5 ppm. Examination of the entire data set revealed that the water test kit gave nine correct results and 10 incorrect results. The Fisher's Test showed that results from the two sets of data were statistically different. The lack of correlation indicates that for the entire data set the technology's results were not accurate. These findings did not change when the entire data set was divided by the site where the samples were collected. The technology produced six correct results and four incorrect results for the former Koppers site data and two correct results and four incorrect results for the Winona Post site data. The other three samples were the PE samples.

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In a comparison of the technology's sample results for the entire data set to the confirmatory laboratory's results, 9 of 19 times the technology was correct. This is 47 percent of the time. Of the other 10 times, the technology gave eight false positive results and two false negative results. This equates to a 42 percent false positive rate and an 11 percent false negative rate. When the data from water samples collected at the former Koppers site is examined alone, 6 of 10 times the technology was correct. This is 60 percent of the time. Of the other four times, the technology gave four false positive results and no false negative results. This equates to a 40 percent false positive rate and a 0 percent false negative rate. When the data from samples collected at the Winona Post site was examined alone, two of six times the technology was correct. This is 33 percent of the time. Of the other four times, the technology gave two false positive results and two false negative results. This equates to a 33 percent false positive rate and a 33 percent false negative rate.

Overall, for the water matrix the test system is conservative. It is not always accurate, relative to the confirmatory laboratory. The lack of correlation to the confirmatory laboratory indicates that this technology does not produce Level 3 data. Based on the developer's QA requirements for the technology's use, this technology can produce Level 2 data. However, the technology never met its developer's specifications for percentages of correct, false positive, and false negative results. These criteria are: (1) less than 12 percent false positive results, (2) less than 1 percent false negative results, and (3) greater than 88 percent correct results. Exceeding the developer's false positive and false negative rates could indicate that this technology is not appropriate for site characterization activities unless a

high percentage of samples are analyzed by a confirmatory laboratory. This technology only exceeded the developer's specifications for false negative frequency for the Winona Post site samples. This indicates that, if this technology is used to assist removal actions at a PCP site where diesel fuel is the PCP carrier, all negative results should be submitted for confirmatory analysis. The failure of the test system to meet its manufacturer's accuracy criteria and its lack of correlation to confirmatory data places this technology into a Level 1 data category.

The technology produced almost twice as high a percentage of correct results for the former Koppers site samples relative to the samples from the Winona Post site. This indicates a trend for greater accuracy for detecting PCP in water when isopropyl ether and butane are the PCP carrier solvents. The two data sets had similar false positive results while the former Koppers site samples produced no false negatives and the Winona Post site samples produced 33 percent false negatives.

### ***Precision***

The precision of the technology when compared to that of the confirmatory laboratory had seven out of 14 soil duplicate pairs agree with the confirmatory laboratory's corresponding duplicate pairs. This equates to a 50 percent precision. This is an unacceptable precision. A similar comparison for the water duplicates showed that two out of six duplicate pairs matched. This equates to a 33 percent precision for the water sample field duplicates. This is an unacceptable precision.

## Section 7

### Ohmicron Corporation: Penta RaPID Assay

This section provides information on the Penta RaPID Assay including background information, operational characteristics, performance factors, a data quality assessment, and a comparison of its results with those of the confirmatory laboratory.

#### Theory of Operation and Background Information

Ohmicron has developed the assay kit to determine PCP concentrations in water, soil, crops, and food. The water and soil applications of the assay kit were evaluated during this demonstration (See Exhibit 7-I). The assay kit uses the principles of ELISA to determine PCP concentrations in water and soil samples. A review of the principles of ELISA is presented in Section 6. The differences in ELISA technology between this kit and the one discussed in Section 6 are presented below.

The developer covalently binds its antibodies to magnetic particles for use. The developer claims there are three advantages to the use of magnetic particles over covalently binding the antibodies to the walls of a test tube. The first is that the magnetic particles are easier to coat and manufacture than the test tubes. The second is that less of the antibody is required for coating the magnetic particles than for the test tubes. The third advantage is that the magnetic particles provide more surface area creating more opportunities for binding.

In addition to the antibodies, ELISA-based technologies use an enzyme conjugate in the analysis step of the immunoassay test. The enzyme conjugate is formed by covalently binding a PCP analog to a horseradish peroxidase enzyme. In the case of the RaPID assay, this enzyme conjugate competes with PCP in an environmental sample for antibody binding sites on the magnetic particles. The enzyme conjugate provides the means for identification and quantitation of PCP. ELISA-based technologies use chromogenic reagents that react specifically with the enzyme conjugate to perform identification and quantitation of PCP.

In the case of the assay kit, the enzyme substrate, hydrogen peroxide, and the chromogen, 3,3',5,5'-tetramethylbenzidine react with the enzyme conjugate to produce a brilliant blue color. Because an exact number of antibody binding sites are used with each sample and an exact number of enzyme conjugate molecules are introduced into each sample, the only variable which exists is the number of PCP molecules present in the environmental sample. The PCP in the sample will compete with the enzyme conjugate molecules for antibody binding sites, thus reducing the amount of blue color formed by the reaction. It is the amount of blue color formed by the sample that is used for identification and quantitation of PCP.

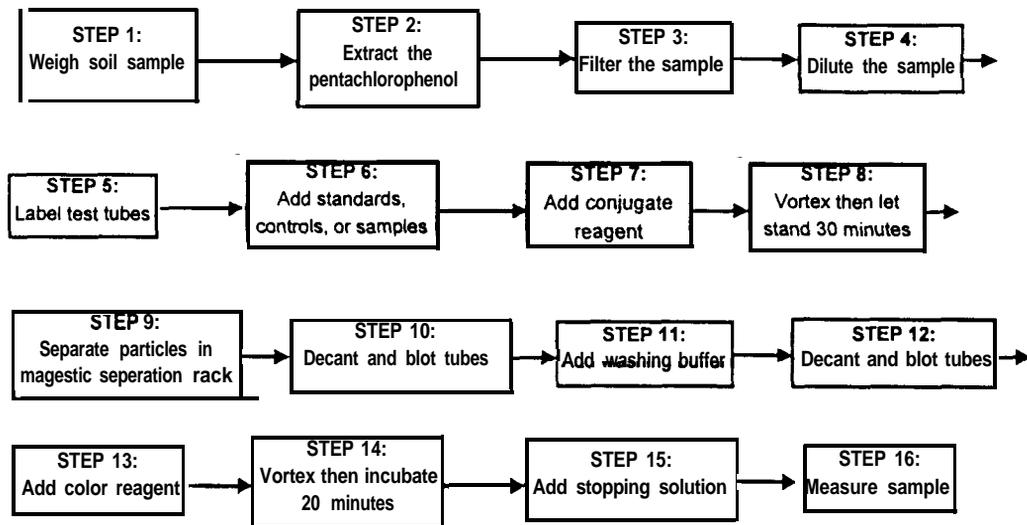
The samples are quantified using the RPA-I RaPID analyzer. PCP results are directly reported in parts per billion for water samples and parts per million for soil samples. The RPA-I RaPID analyzer is an internally calibrated spectrophotometer, operating at a frequency of 450 nanometers. It is equipped with an electronic integrator that has been programmed to perform the three-point standard calibration required for the assay kit. The integrator also has been programmed to flag any calibration QC criteria that are not acceptable.

The assay kit is designed to detect PCP, but other compounds may respond to it as well. This is referred to as cross reactivity, and it is measured by determining the concentration of a compound needed to yield a positive result. The developer has evaluated a number of compounds and has provided information on cross reactivities. The developer suggests that all positive results should be confirmed through the use of an independent, nonimmunological method.

#### Operational Characteristics

The Penta RaPID Assay is composed of three kits: the PCP RaPID Assay Kit, used for both soil and water samples; the Soil Collection Kit, used for soil samples; and the PCP Extraction Kit, used for soil samples.

## OHMICRON CORPORATION PENTACHLOROPHENOL RaPID ASSAY SOIL PROCEDURE



Instrumentation required is shipped in a suitcase-size, plastic container. These containers have small handles and are easy to carry. The reagents and supplies required are shipped in large cardboard boxes. The containers and boxes needed for analysis of 200 samples would fit in the trunk of a large car or in the back seat of a smaller car. The containers and boxes also can easily be shipped by commercial carriers. During this demonstration, all containers and boxes were shipped to the site directly from the developer.

The PCP RaPID Assay Kit can be purchased in either a 30-sample size or a 100-sample size. The following are contained in the assay kit: (1) one bottle of PCP antibody coupled paramagnetic particles (20-milliliter bottle in 30-sample kit, 65-milliliter bottle in 100-sample kit), (2) one bottle of PCP enzyme conjugate (10-milliliter bottle in 30-sample kit, 35-milliliter bottle in 100-sample kit), (3) three vials of PCP standards: 0.1, 2.0, and 10 ppb (2 milliliters each), (4) one vial of a PCP control standard: 1.0 ppb (2-milliliter), (5) one bottle of diluent/zero standard (10-milliliter bottle in the 30-sample kit, 35-milliliter bottle in the 100-sample kit), (6) one bottle of peroxide solution (10-milliliter bottle in the 30-sample kit, 35-milliliter bottle in the 100-sample kit), (7) one bottle of chromogen solution (10-milliliter bottle in the 30-sample kit, 35-milliliter bottle in the 100-sample kit), (8) one bottle of stop solution (20-milliliter bottle in the 30-sample kit, one 25-milliliter and one 40-

milliliter bottle in 100-sample kit), (9) one bottle of washing buffer (70-milliliter bottle in the 30-sample kit, 250-milliliter bottle in the 100-sample kit), (10) polystyrene test tubes (1 box of 36 in the 30-sample kit, 3 boxes of 36 in the 100-sample kit), (11) one copy of instructions, and (12) two sheets of graph paper. The Soil Collection Kit contains the following: (1) twenty-one soil collection devices with detachable plungers and screw caps, (2) twenty-one screw caps with filters, (3) twenty-one extract collection vials, (4) one Styrofoam tube holder, (5) one base piece for the soil collection devices, (6) one copy of the instructions for use of the kit, and (7) forty-five chain-of-custody labels. The PCP Extraction Kit contains the following: (1) twenty bottles of PCP extract solution (20-milliliter each), (2) twenty bottles of PCP extract diluent (25-milliliter each), (3) one 50-microliter, disposable precision pipet, (4) twenty disposable pipet tips, and (5) thirty chain-of-custody labels.

In addition to the above-mentioned kits, other equipment is required for the assays and can be obtained separately from the developer. These items include an RPA-I RaPID Analyzer, a magnetic separation unit, a vortex mixer, a digital balance, a repeating pipet capable of delivering 250, 500, and 1,000 microliters, a 200-microliter fixed volume pipet, and a digital timer. Other equipment which is helpful when using the assays and which is not supplied by the developer includes protec-

tive gloves, twenty-milliliter vials with screw caps for diluting samples, a 10-microliter fixed volume pipet, a pen, and both liquid and solid waste containers.

During the demonstration, the technology was operated in a 28-foot trailer. Electricity was supplied to the trailer for air conditioning and to provide lighting. Electricity also was required to operate the RPA-I RaPID Analyzer during this demonstration. The electricity required was a 110-volt circuit. A refrigerator was required to store analytical reagents for the assay. The developer recommends storing reagents at 2 to 8 °C and has stated that reagents should not be stored at or below 0 °C. A 3-foot by 2-foot hood was used by the operator for soil sample extractions. A 4-foot table was used to perform the assay steps, sample dilutions, sample analysis using the RPA-I RaPID Analyzer, logbook entries, and other data documentation.

The operator of the assay kit was Mr. Nathan Meyer, an employee of PRC with a bachelor of arts degree in biology and a master's of science in environmental science. While at PRC, Mr. Meyer has conducted preliminary site assessments and investigations at hazardous waste sites in EPA Region 7. He also has assisted in RCRA compliance evaluation inspections and in other RCRA enforcement oversight activities. Mr. Meyer's training in the use of the assay kit included viewing a videotape produced by the developer which explained the equipment and provided step-by-step instructions, and reviewing literature provided by the developer. Mr. Meyer also received approximately 8 hours of training at the start of the demonstration from Dr. Scott Jourdon of Ohmicron. This included training in the procedures for extracting, preparing, and analyzing soil and water samples using the assay kit and instructions for use of the pipets and instrumentation. QC procedures and requirements also were discussed. Mr. Meyer then analyzed both soil and water samples using the kit while under the supervision of Mr. Jourdon. After analyzing these samples, Mr. Meyer noted that he felt comfortable with his ability to properly analyze samples.

The assay kit is designed to be operated by persons with some understanding of laboratory procedures. Mr. Meyer had worked in a laboratory and was familiar with laboratory procedures. Mr. Meyer noted that he found the assay kit and its components easy to use, but he also noted that certain steps were tedious and required concentration and consistent technique. The techniques required can be acquired within one week's use of the assay kit. As shown during this demonstration, a person with no experience using the assay kit can produce results. However, the best results can be expected from operators familiar with the kit and have used it before.

The assay kit is promoted for use as a portable field instrument, but does require special care and handling in the field to avoid damage. The RPA-I RaPID Analyzer is a spectrophotometer and electronic spectrophotometric integrator and requires care during shipping and transportation to avoid breaking its internal components. It also requires protection from the elements, such as moisture, sunlight, and extremes of temperature. The balance and pipets are precise measuring instruments and need to be handled with care. No mechanical or electronic problems were experienced during the demonstration.

Instrument reliability was evaluated by monitoring specific calibration and QC checks. Calibration QC criteria for the assay kit include the following: (1) meeting or exceeding a correlation coefficient of 0.990, and (2) obtaining a percent coefficient of variation for calibration standard replicates of less than or equal to 10 percent. The assay kit also includes a PCP control standard. This standard is analyzed with each calibration. The true value of the control standard is 1.0 ppb and the assay kit must determine a value within the range of 0.7 to 1.3 ppb or the calibration is not acceptable. If any of these criteria are not met, sample results cannot be considered valid.

During the demonstration 18 batches of samples were analyzed. This required 18 calibrations. Fourteen of the 18 calibrations met the criteria mentioned above. The first calibration was unacceptable due to a 33.8 percent coefficient of variation for the 2.0 ppb calibrator. The third calibration was unacceptable because the control sample fell outside the acceptable range. The fourth calibration was unacceptable due to a 35.6 percent coefficient of variation for the 2.0 ppb calibrator. The tenth calibration was also unacceptable because both control samples were outside the acceptance range. After three of the first four calibrations were found to be unacceptable, the operator contacted the developer for technical assistance. Ohmicron officials suggested studying the response values for calibrators that exceeded the 10 percent coefficient of variation criteria, then determining which of the two calibrators most closely matched the response value of previous calibrators which met this criterion. The calibration would then be repeated using the calibrator which most closely matched previous acceptable response values. This calibration would then be analyzed twice. It was believed that this would bring the percent coefficient of variation to an acceptable level. The developer also suggested analyzing two aliquots of the control calibrator: one after the acceptable calibration, but before sample analysis, and one at the end of sample analysis. One of the standards must fall within the acceptance range to consider the results valid.

All of the samples analyzed during an unacceptable calibration were reanalyzed during an acceptable calibration, with the exception of sample 026. This sample was analyzed and reported during the unacceptable third calibration. The analysis of this sample was never repeated during an acceptable calibration. The result for sample 026 was not detected above the soil quantitation limit. This was discovered during a technical review of the data. The technical review advised reporting the result, rather than excluding it from data comparison. The reason for this decision was that the QC criterion which was not met, less than or equal to 10 percent coefficient of variation, should not affect a result of this sort.

Overall, PRC found the instrumentation required for the assay kit to be reliable. Immunoassays are enzymatic reactions and can be effected by changes in ambient temperatures. The assay kit may have been affected by the temperature in the trailer during this demonstration. The operator of the assay kit noted several times during the course of sample analysis that QA/QC criteria were most frequently violated in the afternoon. Although PRC did not record temperatures in the trailer during the demonstration, the technology operators noticed a temperature increase in the trailer during the afternoon. The operator of this technology noted that he believed the temperature in the trailer increased 5 to 10 °F in the afternoon, even while the trailer was air conditioned. This temperature fluctuation seemed to affect the standards and QC criteria. The developer stated that increased temperature can increase PCP binding in the substrate, raising reported PCP concentrations. Therefore, temperature fluctuation could cause calibrators and controls to fail QC criteria set at cooler morning times. Operation of this technology in a temperature-controlled environment could eliminate this problem.

The assay kit contains chemicals in quantities ranging from a few milliliter to 500 milliliters. The more dangerous chemicals include methanol, sodium hydroxide, and sulfuric acid. The kit should not be used near ignition points or open flames and care should be taken to avoid dermal, respiratory, and oral contact with methanol. Also; sodium hydroxide, a strong base, and sulfuric acid, a strong acid, should never be mixed due to the strong reaction that will occur. Other chemicals, including PCP in standards, are used in much smaller amounts.

Costs associated with using the assay kit include the costs of the analyzer and reagents, logistical requirements, the operator, and waste disposal. The developer offers two different sizes of kits, a 30-tube kit and a 100-tube kit. These assay kits contain all of the neces-

sary reagents needed to perform water sample analysis and are also required for soil sample analysis. Prices for the 30-tube kit are \$200 per kit for the purchase of one to three and \$180 per kit for the purchase of four or more. Prices for the 100-tube kit are \$450 per kit for the purchase of one to five kits, \$425 per kit for the purchase of six to 10, and \$400 per kit for the purchase of 11 or more.

Soil samples require the purchase of the above-mentioned kit, as well as the Soil Collection Kit and the PCP Extraction Kit. The Soil Collection Kit contains the plasticware and filtration devices required for soil extraction, and the PCP Extraction Kit contains the reagents and pipets needed for soil extraction. Both of these kits contain enough supplies to perform 20 soil sample extractions. The price of the Soil Collection Kit is \$100, and the price of the PCP Extraction Kit is \$127. Ohmicron also offers additional 100-milliliter bottles of PCP assay and extract diluent for \$10. These diluents are needed to perform the immunoassay and to perform dilution of soil sample extracts.

Some of the equipment needed to analyze samples is sold separately. The repeating pipet is offered for \$340, and pipet tips are \$105 for a pack of 100. The 100-microliter, fixed-volume pipet is offered for \$162, and pipet tips are available for \$57 for 10 racks of 96 pipet tips. The developer also offers the 200-microliter, fixed-volume pipet and the 250-microliter, fixed-volume pipet for \$162 per pipet. An alternative to the purchase of these pipets is the purchase of a trivolume pipet which can be adjusted to 100, 200, and 250 microliters, offered for \$260. Pipet tips for the 200- and 250-microliter fixed-volume pipets and for the trivolume pipet are available for \$65 for 10 racks of 100 pipet tips.

The developer offers two different magnetic separation racks for PCP analysis. A 60-position, bench-top separator is available for \$405, and a five-position field kit separator is available for \$130. The 60-position, bench-top separator was used for sample analysis during this demonstration. Other miscellaneous items needed to perform sample analysis are a vortex mixer which costs \$225, a digital timer which costs approximately \$28, and 500 test tubes which cost approximately \$16.50. The RPA-I RaPID Analyzer is sold by the developer and costs \$3,985. Additional rolls of paper for the analyzer are \$6 per roll.

According to the developer, the shelf-life of the reagents used for the assay kit is 1 year from the date of manufacture. An expiration date is printed on each lot of reagents produced. If kits expire within 90 days of the date of purchase, the developer will replace any unused reagents at no charge.

Logistical requirements include an electrical supply capable of operating the instrumentation, a refrigerator, and temperature-control equipment. Costs associated with these requirements may include trailer rental, electrical hookup fees, refrigerator rental or purchase, and electrical or gas usage.

Operator costs will vary depending on the technical knowledge of the operator. As discussed earlier, the kit can be used by individuals with some understanding of laboratory techniques and a minimal amount of technical training, thereby decreasing this cost. Waste disposal is another operating cost. During this demonstration, about 200 samples were analyzed using the analyzer. The waste generated by these analyses filled half a 55-gallon drum. The cost for disposal of one drum of this waste is estimated at \$1,000.

## Performance Factors

The following paragraphs describe the assay kit's detection limits and sensitivities, throughput, linear range, and drift. Specificity is discussed separately due to its complexity.

PRC conducted three test runs in which water samples were analyzed. In all three test runs, the absorbance of the low calibrator divided by the absorbance of the 0.0 ppm calibrator (B/Bo ratios) were less than the 0.90 level recommended by Ohmicron. Because the B/Bo ratio criteria was met, PRC used the minimum detectable concentration of 0.06 ppb that was specified by the developer as the limit of quantitation for water samples analyzed during this demonstration. This is below the 1.0 ppb MCL for PCP.

Soil sample extracts required a 1 to 1,000 dilution when analyzed with the assay kit. When this dilution factor was multiplied by the minimum detectable concentration, and the units were corrected, the theoretical soil detection limit was 0.06 ppm. PRC conducted 12 test runs in which soil samples were analyzed. Five of the 12 runs provided B/Bo ratios that were greater than the 0.90 level recommended by Ohmicron, with the highest ratio being 0.95. Because the 0.90 B/Bo ratio was not met on all test runs, PRC did not use 0.06 ppm as the minimum detectable concentration for soils. Developer product literature indicates that the quantitation limit for soil samples is 0.10 ppm. PRC used 0.10 ppm for the soil quantitation limit.

The developer reports that about 60 minutes is required to analyze a sample using its assay kit. Up to 26 samples can be prepared at the same time to reduce the average analysis time per sample. The developer claims that a proficient user of the assay kit should be

able to analyze 100 to 150 samples in an 8-hour day. During the demonstration, the 193 samples were analyzed in 90 hours; this equals 2 samples per hour. The operator was able to analyze samples in 60 minutes, as reported by the developer. The average number of samples analyzed in a 10-hour work day was 21. This was less than the 100 to 150 samples reported by the developer. The largest number of samples analyzed in one 10-hour day was 64. The operator noted that sample analysis time did not include the time required for sample handling, data documentation, difficult extractions, or the preparation of QC samples. Samples which exceeded the linearity range required dilution. Seventy-nine samples required at least one dilution, and several of these samples required more than one dilution. The time required by the operator to perform these tasks prevented him from completing analysis of 100 to 150 samples per day.

The linear range of the assay kit is from 0.10 to 10.0 ppb for water samples, and from 0.10 to 10.0 ppm for soil samples. The ranges are established by a three-level calibration using 0.10, 2.00, and 10.0 ppb calibrators. The linear range for the soil samples is defined as the concentration of the calibration standards (low- and high-level calibrators) multiplied by the appropriate factors introduced by the soil extraction. Linearity of the calibration is evaluated using the standard curve of the absorbance versus concentration for each calibration standard level. The developer defines acceptable linearity as a correlation coefficient of 0.990 or greater. Samples which exceed the linear range require dilution to determine the concentration of PCP in the sample. Water samples are diluted by using less of the sample for the assay procedure. This procedure can be employed down to volumes of 10 microliters. When the analysis of 10 microliters exceeds the linear range, the water sample is diluted with deionized water. Soil samples are diluted by performing 1- to 100-fold dilutions of the sample extract. These dilutions are continued until sample results are within the linear range. Several times during the demonstration, a sample which was found to be above the linear range was diluted and reanalyzed. If the result for the diluted sample was still above the upper linear range, the sample extract was diluted further and reanalyzed.

Drift normally is a measurement of an instrument's variability in quantitating a known amount of a standard. The assay kit eliminates the variability associated with drift by requiring a new calibration with each batch of samples analyzed. The absorbance values from the standards analyzed were found to drift during the 18 calibrations performed. For example, the absorbance values obtained from the 0.10 ppb calibrator ranged from 0.792 to 1.656. This is a significant range and

gives justification to the requirement of performing a new three-level calibration for each set of samples analyzed.

### Specificity

Specificity refers to a technology's ability to identify and quantitate a particular contaminant in the presence of chemicals that could act as interferants. For this technology, interferants might be natural chemicals present in a matrix, carriers or other chemicals used to introduce PCP during wood treatment, or other contaminants that might be present at such facilities. To assess the specificity, PRC studied the assay's chemistry, reviewed its developer's literature, and conducted a specificity study.

Organic sample matrix effects are primarily caused by humic acids found in highest concentrations in topsoil. Humic acids can leach from soil and, therefore, can be found in water samples, as well. High concentrations of humic acids in a sample can cause a loss of recovery of PCP through absorption. The assay kit uses a methanol and sodium hydroxide buffer solution for sample extraction and assay diluent to eliminate the absorption of PCP by humic acids. The developer claims that humic acids up to 10 ppm have no effect on the assay kit. Inorganic sample matrix effects are primarily caused by pH, salinity, and inorganic chemical composition. The immunoassay portion of the analysis must be performed under basic conditions. The developer recommends that samples with a low pH be neutralized with a 6 Normal sodium hydroxide solution before performing the assay. The salinity of water may sometimes affect immunoassay results. The developer reports that the assay kit is not affected by solutions containing up to 0.65 Molar sodium chloride.

Some inorganic chemicals may have an effect on the performance of the assay kit. Inorganic compounds may inhibit the binding of PCP or the enzyme conjugate to the antibody binding sites. The developer has evaluated a number of inorganic compounds which may affect the performance of the kit. These chemicals and their tested concentrations, as reported by the developer, are shown in Table 7- 1.

One objective of this demonstration was to evaluate the technology's ability to quantitate PCP concentrations in samples contaminated with diesel fuel and isopropyl ether, which were used as carriers to introduce the PCP

into wood at the two wood treatment facilities. Ohmicron has evaluated some solvents similar to the isopropyl ether for their effect on the assay and found that the minimum concentration needed to affect the

TABLE 7-1. INORGANIC CHEMICAL RESPONSE AS REPORTED BY OHMICRON

Compound	Concentration' Tested (ppb)
Calcium	250,000
Copper	250,000
Iron	50,000
Manganese	250,000
Magnesium	250,000
Mercury II	250,000
Nickel	250,000
Nitrate	250,000
Phosphate	250,000
Sodium chloride	650,000
Sulfate	10,000,000
Sulfite	250,000
Thiosulfate	250,000
Zinc	250,000

Note:

a No response was found at these concentrations.

results is 5 percent acetone, 2 percent acetonitrile, and 10 percent methanol. Because isopropyl ether has a similar chemical structure to these solvents, the maximum concentration before an effect is produced should be similar. Percent levels of isopropyl ether or butane were not expected in the samples from the former Koppers site, due to their volatility and relatively short residence time in surficial soils. It is possible that percent levels of diesel fuel were encountered in the samples from the Wiiona Post site, but the developer reports that diesel fuel concentrations up to 10 percent will not affect the kit. In addition to these PCP carriers, other wood preserving agents are often used in conjunction with PCP. The developer has evaluated two of these wood preserving agents and determined that the kit is not affected by concentrations of 100 ppm creosote or 1,000 ppm CCA.

Another site-specific matrix effect is the presence of other chemicals present in the samples. Historical data for the two demonstration sites revealed the presence of other phenols, including chlorophenols, as well as dioxins and furans, particularly the octa-isomers. The effects of the nonchlorinated phenols, dioxins, and furans were not evaluated during this demonstration, nor was any information regarding their effects provided by the developer. However, the effects of these chemicals on the assay kit are believed to be insignificant compared to those of the chlorophenols, due to their lesser chemical similarity to PCP.

As shown in Table 7-2, chemicals that have very similar chemical structure to PCP (trichlorophenols and tetrachlorophenols) show a high degree of cross

TABLE 7-2. COMPOUND CROSS REACTIVITY AS REPORTED BY OHMICRON

Compound	Concentration Needed to Provide a Positive Result (ppb)
Pentachlorophenol	0.66
2,3,5,6-Tetrachlorophenol	0.21
2,3,4,6,-Tetrachlorophenol	0.91
2,3,5-Trichlorophenol	1.52
2,3,6-Trichlorophenol	2.44
Tetrachlorohydroquinone	8.70
2,4,6-Trichlorophenol	15.1
2,4,5-Trichlorophenol	21.5
2,3,4-Trichlorophenol	53.2
2,5-Dichlorophenol	62.9
2,6-Dichlorophenol	286
2,3-Dichlorophenol	611
2,4-Dichlorophenol	887
3,5-Dichlorophenol	1,670
Hexachlorobenzene	1,560
Hexachlorocyclohexane	5,790

reactivity; other chemicals, including the dichlorophenols are less chemically similar to PCP, and have a very low cross reactivity, less than 0.0001 of the response of PCP.

Ohmicron also has tested the following chemicals at a level of 10,000 ppb and found they did not give a positive result using the assay:alachlor, aldicarb, benomyl, butachlor, butylate, captan, carbaryl, carben-dazim, carbofuran, 4-chlorophenol, chlorthalonil, 2-4-D, 3,4-dichlorophenol, 1,3-dichloropropane, dinoseb, matelaxyl, metalochlor, metribuzen, pentachlorobenzene, pentachloronitrobenzene, picloram, propachlor, terbos, triclopyr, thiobendazole, and thiphenate-methyl. Many of these chemicals contain an aromatic ring group similar to PCP; the antibody lock appears to be geared for the hydroxyl group of the PCP key.

The specificity study involved spiking clean sand with known concentrations of chlorophenols and diesel fuel. The soil and water specificity samples were prepared by the lead chemist and given to the technology operator along with the demonstration samples. The soil specificity samples were prepared by weighing 10 grams of clean sand, placing it into a soil extraction device, and spiking it with microliter amounts of the chemical standards. One soil specificity sample, SS-17, was an unspiked sand sample. It did not give a positive result.

Soil specificity samples SS-01 through SS-04 were spiked with 50 ppm of diesel fuel. These samples did

not give a positive result. Soil specificity samples SS-09 through SS-12 were spiked with 5 ppm of 2,4-dichlorophenol. These samples did not give a positive result. Soil specificity samples SS-13 through SS-16 were spiked with 5 ppm of 2,4,6-trichlorophenol. Two of these samples did not give positive results. Two of the samples, though, did give positive results. The result of samples SS-13 and SS-15 were 0.11 ppm and 0.06 ppm, respectively. The result for SS-15 was below the soil quantitation limit, and it would be reported as not detected during a field sampling event. Still, the developer states that the cross reactivity of 2,4,6-trichlorophenol is 15.1 ppm. The results of the specificity study may indicate that the actual cross reactivity of this compound is lower than this level. Soil specificity samples SS-05 through SS-08 were spiked with 5 ppm of 2,3,4,6-tetrachlorophenol. All of these samples produced a positive result. Sample results ranged from 0.32 ppm to 0.53 ppm. The mean result from the four samples was 0.41 ppm. The developer claims that the cross reactivity of 2,3,4,6-tetrachlorophenol is 0.91 ppm. The specificity test results show that this claim is accurate.

The water specificity samples, SS-18 through SS-21, were spiked with 125 ppm of diesel fuel and 50 ppb of PCP. This was done to evaluate the effects of diesel fuel on the recovery of PCP in water-samples. The water specificity samples ranged in concentration from 42 to 70 ppb, with a mean result of 57.8 ppb. Recoveries for PCP ranged from 84 to 140 percent, with a mean recovery of 116 percent. If these values are compared with the matrix spike values, the mean recovery of the water specificity samples is equivalent to the water matrix spike recoveries obtained. Based on this comparison, diesel fuel at 125 ppm does not interfere with the recovery of PCP at a concentration of 50 ppb.

### Intramethod Assessment

Intramethod measures of the technology's performance included its results on reagent blanks, the completeness of its results, its intramethod accuracy, and its intramethod precision. Reagent blank samples were prepared by taking reagents through all extraction, cleanup, and reaction steps of the analysis. Soil reagent blank samples were extraction solvent added to the extraction tubes, while the water reagent blank sample was deionized water added to the assay tube. An acceptable reagent blank sample must not contain PCP above the method quantitation limit. Eleven reagent blanks were analyzed during the demonstration. Ten of the eleven reagent blanks were found to contain no PCP above the method quantitation limit and were determined to be acceptable.

TABLE 7-3. PENTA RaPID ASSAY PCP PROFICIENCY SAMPLE RESULTS: Training Results

Sample No.	Result (ppb)	Mean Result (ppb)	Percent Recovery (%)	± 2 Standard Deviation Range (ppb)	Did Result Fall Within Acceptance Range (Yes or No)
A	0.63	0.64	98	0.40 to 0.87	Yes
B	1.87	2.63	71	1.87 to 3.40	Yes
C	4.22	5.12	82	3.33 to 6.90	Yes

TABLE 74. PENTA RaPID ASSAY PCP PROFICIENCY SAMPLE RESULTS: Demonstration Results

Sample No.	Result (ppb)	Mean Result (ppb)	Percent Recovery (%)	± 2 Standard Deviation Range (ppb)	Did Result Fall Within Acceptance Range (Yes or No)
A	0.31	0.64	48	0.40 to 0.87	No
B	1.05	2.63	40	1.87 to 3.40	No
C	3.94	5.12	77	3.33 to 6.90	Yes

One reagent blank sample (RB-6) was found to contain 0.13 ppm of PCP. This did not meet the reagent blank requirements. Corrective action required for unacceptable reagent blanks included a detailed data review and, if necessary, the reanalyzing of the reagent blank. If the reagent blank was still unacceptable, corrective action called for the reanalysis of the reagent blank and all samples associated with it. RB-6 was analyzed during test run six, which included 20 soil samples, one laboratory duplicate sample, and a matrix spike and matrix spike duplicate sample. PRC closely reviewed the data from test run 6 to determine whether the unacceptable reagent blank sample affected sample results. Two samples were reported as “ND” by the RPA-I RaPID Analyzer. This indicates that the sample contained less than 0.10 ppm, as reported by the Analyzer. Since these samples contained less than the unacceptable reagent blank, PRC feels confident in reporting these samples as not detected above 0.10 ppm. The other samples analyzed during test run six were found to contain PCP. All these sample results, though, were more than five times the amount of PCP found in RB-6. PRC feels confident the reported values for all samples analyzed during test run six were not affected by the unacceptable reagent blank, RB-6. Therefore, no corrective action was taken.

For this demonstration, completeness refers to the proportion of valid, acceptable data generated. Completeness for the samples analyzed by the assay was 100 percent, well above the objective of 90 percent.

Intramethod accuracy was assessed for the technology through the use of proficiency samples, PE samples, and matrix spike and matrix spike duplicate samples.

Proficiency samples were provided by the developer to verify and document operator accuracy. The proficiency samples were analyzed two times, once during the training period under the supervision of the developer representative and once during the demonstration.

The developer has analyzed each of the proficiency samples using its technology and has determined a mean value and the 90 percent confidence interval ( $\pm 2$  standard deviation range) for each of the proficiency samples. As Table 7-3 shows the operator was able to produce results for all three of the proficiency samples which were within this range. These results show that the operator was proficient in the use of the assay kit prior to the start of the demonstration. Table 7-4 shows the results for the proficiency samples analyzed during the demonstration. Two of the three sample results were outside of the  $\pm 2$  standard deviation range recommended by the developer. In both samples, the results were lower than expected. Recoveries of these two samples compared to the mean value of the proficiency sample were 48 and 40 percent. Overall, 67 percent of the proficiency samples analyzed were within the  $\pm 2$  standard deviation range deemed by the developer to be acceptable. The results of these proficiency samples may indicate that field personnel recently trained in the use of the assay kit may not be able to continuously produce results which are as accurate as those produced by more experienced personnel.

Five PE samples were analyzed during the demonstration, two for soil and three for water. Both of the soil PE samples and two of the water PE samples were purchased from ERA; the other water PE sample was produced by PRC. These samples were extracted

**TABLE 7-5. PENTA RaPID ASSAY SOIL PERFORMANCE EVALUATION SAMPLE RESULTS**

Sample No.	Result (ppm)	True Result (ppm)	Percent Recovery %	Acceptance Range (ppm)	Did Result Fall Within Acceptance Range (Yes or No)
099	5.01	7.44	67	1.1 to 13	Yes
100	38	101	38	15 to 177	Yes

**TABLE 7-6. PENTA RaPID ASSAY WATER PERFORMANCE EVALUATION SAMPLE RESULTS**

Sample No.	Result (ppb)	True Result (ppb)	Percent Recovery (%)	Acceptance Range (ppb)	Did Result Fall Within Acceptance Range (Yes or No)
196	62	68.4	91	10 to 120	Yes
107	1,100	2,510	44	377 to 4,420	Yes
113	7.62	7.50	102	2.25 to 12.8	Yes

**TABLE 7-7. PENTA RaPID ASSAY SOIL MATRIX SPIKE SAMPLE RESULTS**

Sample No.	Amount Found In Original Sample and Duplicate Sample (ppm)	Amount Added To Matrix Spike Sample (ppm)	Amount Found In Matrix Spike Sample (ppm)	Sample Percent Recovery (%)	Amount Found In Matrix Spike Duplicate Sample (ppm)	Duplicate Percent Recovery (%)	Relative Percent Difference (%)
020	0.13	5.00	3.60	69	4.21	82	17
029	ND	5.00	4.57	91	4.65	93	2
039	ND	5.00	5.56	110	6.23	125	13
051	ND	5.00	5.20	164	4.74	95	5
086	1.86	5.00	8.46	132	8.68	136	3
090	ND	5.00	5.87	117	4.95	99	17
995	ND	5.00	6.31	126	4.94	99	24

Note:  
 ND None detected above soil quantitation limit of 0.10 ppm

and analyzed in the same way as the other samples. The operator did not know that the samples were PE samples, nor did the operator know the true concentration and acceptance range. The results for the PE samples are in Tables 7-5 and 7-6 All values reported for the PE samples were within acceptance ranges. Accuracy of the samples analyzed was found to be 100 percent for both sample matrices.

Matrix spike samples are aliquots of original sample into which a known concentration of PCP is added. These samples are then extracted and analyzed in the same way as original samples. Seven soil samples and one water sample were used for matrix spike samples. They also were duplicated. Therefore, 14 spiked soil

samples and two spiked water samples are used in this assessment. The average recovery of the soil matrix spike samples and duplicates was 106 percent or 5.3 ppm. This is very close to the 5 ppm actually added to the samples. The standard deviation of the matrix spike samples was 19 percent or 0.95 ppm. Control limits for soil matrix spike recovery can be established following guidelines outlined in the SW-846 Manual Method 8000. Control limits are defined as  $\pm 2$  standard deviations from the mean. For the soil matrix spike samples analyzed during the demonstration, the calculated control limits ranged from 68 to 144 percent recovery. All soil matrix spike samples analyzed fell within these control limits. The soil matrix spike results are shown on Table 7-7. The recoveries for the water matrix spike sample

**TABLE 7-8. PENTA RaPID ASSAY LABORATORY DUPLICATE RESULTS: Soil Samples**

Sample No.	Original Sample Result (ppm)	Laboratory Duplicate Result (ppm)	Sample	Relative Percent Difference (%)
004	0.65	0.75		14
018	2.77	2.02		31
019	3.90	3.78		3
038	4.61	2.24		69
057	7.72	3.09		86
058	2.92	3.01		3
088	1.51	1.19		24
099	5.01	2.86		55

and its duplicate were 116 and 115 percent, respectively. Their relative percent difference was 1 percent. Because only two recoveries were produced for water matrix spike samples, control limits similar to those developed with the soil matrix spike samples were not established.

Intramethod precision for the kit was assessed by comparing the results obtained on duplicate samples. Three types of precision data were generated: data from laboratory duplicate samples, data from field duplicate samples, and data from matrix spike duplicate samples. Usually these duplicate samples are used to determine matrix variability and the effects of using several operators. To use the duplicates to measure the method's precision, PRC both controlled for matrix variability by thoroughly homogenizing the samples and controlled for operator effects by using only one operator for the entire demonstration. Results for soil laboratory duplicate samples are provided in Table 7-8. Soil laboratory duplicate samples were analyzed after the original sample results were obtained. Only samples with positive results were used for laboratory duplicate analysis. Eight soil laboratory duplicate samples were analyzed during the demonstration. The original results obtained for these samples ranged from 0.65 to 7.72 ppm. When the analysis was duplicated, the results ranged from 0.75 to 3.78 ppm. RPD values for the soil laboratory duplicate samples ranged from 3 to 86 percent. The mean RPD value of the soil laboratory duplicate samples was 36 percent, with a standard deviation of 31 percent.

Water laboratory duplicate samples were analyzed after the original sample results were obtained. Only samples with positive results were used for laboratory duplicate analysis. Two water laboratory duplicate samples were analyzed. The original results obtained for these samples were 4.34 and 1.42 ppb. When the analysis was duplicated, the results were 4.34 and 1.43

ppb. RPD values for the water laboratory duplicate samples were 0 and 1 percent.

Fourteen field duplicate soil samples were analyzed. In one, the kit did not detect PCP. This sample was eliminated from the statistical analysis. The remaining results obtained for the soil field duplicate samples ranged from 1.17 to 11,800 ppm. The field duplicate sample results ranged from 1.46 to 11,700 ppm. RPD values for the soil field duplicate samples ranged from 1 to 118 percent. The mean RPD value these samples was 28 percent, with a standard deviation of 31 percent. The results are shown on Table 7-9. Ten water field duplicate samples were analyzed. Field duplicate samples represent 43 percent of all water samples analyzed during the demonstration. The original results obtained for the water samples ranged from 0.71 to 122,000 ppb. The field duplicates results were from 0.94 to 95,600 ppb. RPD values for the water field duplicate samples ranged from 1 to 34 percent. The mean RPD value of the water field duplicate samples was 20 percent, with a standard deviation of 9 percent. These results are shown on Table 7-10.

PRC used the laboratory and field duplicates together to evaluate the technology's precision. To do this, PRC established control limits like those sometimes used to evaluate laboratory duplicates. These control limits were then used to determine whether the difference between a result from a duplicate and the result from its respective sample was reasonable. To establish the control limits, all sample pairs that did not produce two positive results were removed from the data population. Then, the RPD for each pair was calculated and the mean RPD and population standard deviation were determined. The lower control limit was set at zero because this would mean that the results from a duplicate and its sample matched perfectly. The upper

**TABLE 7-9. PENTA RaPID ASSAY SOIL FIELD DUPLICATE SAMPLE RESULTS**

Sample No.	Original Sample Result (ppm)	Field Duplicate Sample Result (ppm)	Relative Percent Difference (%)
001	1.61	2.53	44
011	55	64	15
020	0.13	ND	NA
030	8.3	8.6	3
040	19	10	62
048	11,800	11,700	1
050	1.17	1.46	22
055	748	670	11
058	2.92	2.16	30
059	1,800	2,200	20
073	123	125	2
074	649	690	6
086	1.86	7.21	118
087	29	20.2	36

**Notes:**

- ND None detected above soil quantitation limit of 0.10 ppm.
- NA Not applicable due to ND result.

control was set by multiplying the standard deviation by two and adding it to the mean RPD. The RPD of each sample pair was then compared to these control limits. If greater than 90 percent fell within the control limits, the technology's precision was considered adequate. If fewer than 90 percent of them fell within this range, the data was reviewed, and if no explanation could be found, the technology's precision was considered inadequate.

The assay kit for soils had 21 duplicates in which both a sample and its duplicate had positive results. The data from these 21 pairs had a mean RPD of 31 percent and a standard deviation of 3.1. The control limits were, therefore, set at 0 and 93 percent. All but one of the 21 RPDs fell within the control limits. The sample pair that was outside the control limits had results of 1.86 and 7.21 ppm, respectively. That sample pair had an RPD of 118 percent. Still, 95.2 percent of the sample pairs had RPDs within the control limits. Based on this, the precision of the assay kit for soil samples was found to be acceptable.

The assay kit for water had 12 duplicate pairs in which both a sample and its duplicate had positive

results. The data from these pairs had a mean RPD of 17 percent and a standard deviation of 11. The control limits were, therefore, set at 0 and 39 percent. All 12 fell within these limits, and the precision for the assay kit for water samples was found to be acceptable.

Matrix spike duplicate samples were used to further evaluate precision. Seven soil matrix spike duplicate samples and one water matrix spike duplicate sample were analyzed and their results were compared to the results of their respective matrix spike samples. Precision of the matrix spike duplicate samples was evaluated through the RPD of the matrix spike result and the matrix spike duplicate result. RPD values for the seven pairs of matrix spike soil samples ranged from 2 to 24 percent. The mean RPD value from these seven pairs was 12 percent, and the standard deviation was 8 percent. If an upper control limit of two times the standard deviation is used, the upper control limit for RPD determined for soil samples analyzed during the demonstration was 28 percent. All RPD values for the soil matrix spike duplicate samples were below the upper control limit. The test kit's precision, therefore, was found to be acceptable. Precision of the water matrix spike duplicate sample was evaluated by examining the

TABLE 7-9. PENTA RaPID ASSAY SOIL FIELD DUPLICATE SAMPLE RESULTS

Sample No.	Original Sample Result (ppm)	Field Duplicate Sample Result (ppm)	Relative Percent Difference (%)
001	1.61	2.53	44
011	55	64	15
	0.13	ND	NA
	6.3	8.6	3
	19	10	62
	11,800	11,700	
	1.17	1.46	22
055	748	670	11
058	2.92	2.16	30
059	1,800	2,200	20
	123	125	2
074	649	690	6
086	1.86	7.21	118
087	29	20.2	36

## Notes

- ND None detected above soil quantitation limit of 0.10 ppm.  
 NA Not applicable due to ND result.

control limit was set by multiplying the standard deviation by two and adding it to the mean RPD. The RPD of each sample pair was then compared to these control limits. If greater than 90 percent fell within the control limits, the technology's precision was considered adequate. If fewer than 90 percent of them fell within this range, the data was reviewed, and if no explanation could be found, the technology's precision was considered inadequate.

The assay kit for soils had 21 duplicates in which both a sample and its duplicate had positive results. The data from these 21 pairs had a mean RPD of 31 percent and a standard deviation of 3.1. The control limits were, therefore, set at 0 and 93 percent. All but one of the 21 RPDs fell within the control limits. The sample pair that was outside the control limits had results of 1.86 and 7.21 ppm, respectively. That sample pair had an RPD of 118 percent. Still, 95.2 percent of the sample pairs had RPDs within the control limits. Based on this, the precision of the assay kit for soil samples was found to be acceptable.

The assay kit for water had 12 duplicate pairs in which both a sample and its duplicate had positive

results. The data from these pairs had a mean RPD of 17 percent and a standard deviation of 11. The control limits were, therefore, set at 0 and 39 percent. All 12 RPDs fell within these limits, and the precision for the assay kit for water samples was found to be acceptable.

Matrix spike duplicate samples were used to further evaluate precision. Seven soil matrix spike duplicate samples and one water matrix spike duplicate sample were analyzed and their results were compared to the results of their respective matrix spike samples. Precision of the matrix spike duplicate samples was evaluated through the RPD of the matrix spike result and the matrix spike duplicate result. RPD values for the seven pairs of matrix spike soil samples ranged from 2 to 24 percent. The mean RPD value from these seven pairs was 12 percent, and the standard deviation was 8 percent. If an upper control limit of two times the standard deviation is used, the upper control limit for RPD determined for soil samples analyzed during the demonstration was 28 percent. All RPD values for the soil matrix spike duplicate samples were below the upper control limit. The test kit's precision, therefore, was found to be acceptable. Precision of the water matrix spike duplicate sample was evaluated by examining the

**TABLE 7-10. PENTA RaPID ASSAY WATER FIELD DUPLICATE SAMPLE RESULTS**

Sample No.	Original Sample Result (ppb)	Field Duplicate Sample Result (ppb)	Relative Percent Difference (%)
101	9.00	8.87	1
102	53,000	59,100	11
103	122,000	95,600	24
104	2,300	1,800	24
105	31	40	25
108	4.34	5.01	14
109	1.42	2.00	34
110	30	24	22
111	0.71	0.94	28
112	1,500	1,800	18

RPD between the matrix spike result and the matrix spike duplicate result. The RPD value for the matrix spike water samples was 1 percent.

### Comparison of Results to Confirmatory Results

The quantitative results of the assay kit were compared to those of the confirmatory laboratory using the statistical methods detailed in Section 4 (see Tables 7-11, 7-12, and 7-13). The purpose of this statistical data evaluation is to assess whether the technology meets Level 3 criteria for accuracy and precision. If the technology cannot, but the technology's data can be mathematically corrected to become accurate, it can be placed into a Level 2 data quality category.

The Wilcoxon Signed Ranks Test was used to supplement the findings of the regression analysis. This nonparametric test was used to test the hypothesis that the technology's data was not significantly different from the confirmatory laboratory's data. In cases where the regression analysis criteria for accuracy were not met, but the Wilcoxon Signed Ranks Test indicated that there was no significant difference between the two data sets, the technology's data was placed in the Level 2 category. The contradiction between the regression and inferential statistic indicates that the distribution of one or both of duplicate had positive results. The data from these 12 pairs had a mean RPD of 17 percent and a standard deviation of 11. The control limits were, therefore, set at 0 and 39 percent. All 12 RPDs fell within the control the data sets violated a fundamental assumption of regression analysis, normal data distribution. In these cases, the regression analysis was discarded, and

evaluation was based on the inferential statistic. Data not meeting any of the above criteria was placed into a Level 1 data quality category, unless it was unable to detect PCP when PCP was in a sample. Identifying the presence of a compound is a criterion of Level 1 data. The Wilcoxon probability and the parameters for the regression analysis are presented in Table 7-14.

This technology was assessed in both soil and water. For each of these matrices two different sites were sampled. One site was contaminated with PCP in an isopropyl ether and butane carrier solvent and the other site was contaminated with PCP in a diesel fuel carrier solvent.

The statistical evaluation of this technology was carried out in a tiered approach. For the soil matrix, if the initial analysis of the entire data set (tier 1) showed that the technology's data was statistically different from the confirmatory laboratory's data, the data set was divided by the site producing the samples. This comparison was used to conduct a preliminary assessment of any carrier effect on the technology's data (tier 2). A third tier of analysis was conducted within each site-specific data set. This assessment involved splitting the data sets into two subsets again, one having samples the confirmatory laboratory found had concentrations less than 100 ppm, the other those having concentrations greater than 100 ppm. This assessment was used to address any potential concentration effects on the technology's performance. The 100 ppm level was selected as a general action level for PCP in soil. EPA action levels for this contaminant are generally site specific; however, they typically occur in a range between 20 and 100 ppm.

TABLE 7-1 1. SUMMARY OF DEMONSTRATION DATA FOR FORMER KOPPERS SITE SOIL SAMPLES

Sample No.	Penta RaPID Assay (0.10 ppm)	Confirmatory Laboratory (ppm)	Sample No.	Penta RaPID Assay (0.10 ppm)	Confirmatory Laboratory (ppm)
001	1.61	4.42	031	0.70	1.43
001D	2.53	4.18	032	0.12	0.62 <sup>b</sup>
002	0.81	1.64	033	0.15	0.40
003	<0.10 U	0.13 <sup>b</sup>	034	0.23	0.31 <sup>b</sup>
004	0.65	2.04	035	49.8	145
005	4.82	3.70	036	28.9	36.8
006	0.66	1.89	037	1.54	1.19
007	3.83	2.66	038	4.61	77.0
008	0.32	0.66	039	<0.10 u	3.32
009	0.45	3.52	040	19.0	400.0
010	289	435.0	040D	10.0	34.40
011	55.0	106.0	041	12.0	6.44
011D	64.0	112.0	042	5.39	4.09
012	<0.10 U	0.056 <sup>b</sup>	043	585	655.0
013	17.0	32.80	044	4,000	6,956
014	46.0	99.60	045	13.0	22.10
015	135	1,190	046	1.14	0.95
016	122	273.0	047	32,000	13,920
017	43.2	1,335	048	11,800	26,100
018	2.77	2.13	048D	11,700	30,260
019	3.90	6.89	049	105	255.0
020	0.13	0.10	050	1.17	2.16
020D	<0.10 U	0.09 <sup>b</sup>	050D	1.46	1.25
021	400	5,320	051	<0.10 u	0.43
022	19.8	1.85	052	24.00	28.20
023	9.70	1.86	053	1.49	2.23
024	0.52	1.57	054	<0.10 u	0.47
025	86.0	593	055	748	3,135
026	<0.10 U	0.42	055D	670	3,003
027	3.80	11.3	056	668	9.90
028	0.28	0.45	057	7.72	8.74
029	<0.10 U	1.06	058	2.92	3.53
030	8.30	28.6	058D	2.16	9.13
030D	8.60	29.0			

## Notes:

<sup>a</sup> Detection limits are presented in parentheses.<sup>b</sup> Sample was analyzed by Method 8151A; all other samples were analyzed by Method 8270A.

J Reported amount is below detection limit or not valid by approved QC procedures.

U PCP was not detected.

TABLE 7-12. SUMMARY OF DEMONSTRATION DATA: WINONA POST SOIL SAMPLES

Sample No.	Penta RaPID Assay (0.10 ppm) <sup>a</sup> .	Confirmatory Laboratory (ppm)	Sample No.	Penta RaPID Assay (0.10 ppm) <sup>a</sup> .	Confirmatory Laboratory (ppm)
059	1,800	9,600	080	1,700	2,550
059D	2,200	10,260	081	138	125.0
060	877	1,008	082	1,000	2,400
061	1,840	2,744	083	107	270.0
062	32.0	138.0	084	565	1,140
063	462	1,610	085	67.0	57.70
064	1,030	1,978	086	1.86	6.59
065	604	1,577	086D	7.21	6.88
066	96.0	57.80	087	29.0	34.00
067	52.0	110.0	087D	20.2	51.80
068	19.0	47.70	088	1.51	2.58
069	406	798.0	089	0.16	0.21 <sup>b</sup>
070	766	2,888	090	<0.10 U	0.55 <sup>b</sup>
071	236	289.0	091	0.27	0.28 <sup>b</sup>
072	522	336.0	092	0.31	0.57 <sup>b</sup>
073	123	74.80	093	0.36	0.19 <sup>b</sup>
073D	125	78.20	094	0.27	1.02 <sup>b</sup>
074	649	836.0	095	<0.10 U	0.088 <sup>b</sup>
074D	690	1,520	096	22.0	59.80
075	1,800	3,692	097	10.0	14.60
076	1,800	4,590	098	1.25	0.57
077	1,900	2,040	099	5.01	4.02
078	1,250	1,720	100	38.00	52.40
079	505	792.0			

a Detection limits are presented in parentheses.

b Sample was analyzed by Method 8151A; all other samples were analyzed by Method 8270A.

J Reported amount is below detection limit or not valid by approved QC procedures.

U PCP was not detected.

TABLE 7-1 3. SUMMARY OF DEMONSTRATION DATA: Water Data<sup>a</sup>

Sample No.	Penta RaPID Assay (0.00006 ppm) <sup>b</sup>	Confirmatory Laboratory (ppm)
101	0.00900	0.004140 <sup>c</sup>
102	53.0	15.90
103	122	13.50
104	2.30	0.01230
105	0.031	0.8490
105D	0.040	0.6400
106	0.062	0.01030 <sup>''</sup>
107	1.10	2.050 <sup>c</sup>
108	0.00434	0.001850 <sup>c</sup>
108D	0.00501	0.00221 0 <sup>c</sup>
109	0.00142	0.0001 750 <sup>c</sup>
109D	0.00200	0.0006300 <sup>c</sup>
110	0.0300	0.01810 <sup>''</sup>
110D	0.0240	0.01810 <sup>c</sup>
111	0.000710	0.0003480 <sup>c</sup>
111D	0.000940	0.0003200 <sup>''</sup>
112	1.50	1.810
112D	1.80	2.020
113	0.00762	0.002270 <sup>c</sup>

Notes:

- a Samples 101 through 105D were collected from the Winona Post site. Samples 108 through 112D were from the former Koppers site. Samples 106, 107, and 113 were PE samples.
- b Detection limit.
- J Samples analyzed by Method 515.1; all other samples were analyzed by Method 8270A.
- U Reported amount is below detection limit or not valid by approved QC procedures.
- PCP was not detected.

Precision for this technology was assessed through the use of the Dunnett's Test, and a Wilcoxon Signed Ranks Test. The Dunnett's Test was used to see if the technology's precision was different from the confirmatory laboratory's precision, and the Wilcoxon Signed Ranks Test was used to verify the results of the Dunnett's Test.

### Soil Samples: Accuracy

The initial linear regression analysis of the entire data set was based on results from 90 samples. The other samples did not contain PCPs above the detection limit of 0.10 ppm. The  $r^2$  for this regression was 0.47, indicating that a relationship may exist between the data sets. A residual analysis of the data, though, identified samples 21, 44, 47, 48, 59, and 77 as outliers. PRC removed these six points and recalculated the linear regression. When the regression was recalculated on the 84 remaining sample results, it defined an  $r^2$  of 0.81, indicating that a relationship exists between the two data

FIGURE 7-1. TOTAL SOIL DATA SET

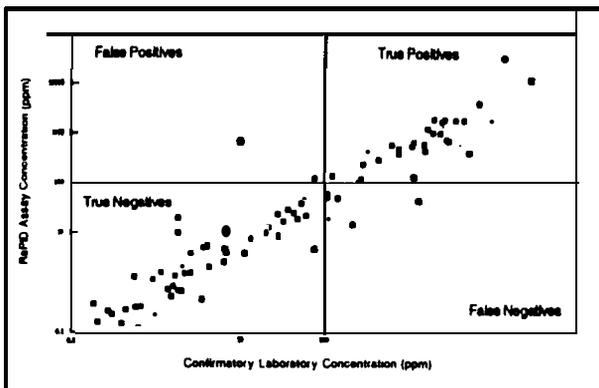


FIGURE 7-2. FORMER KOPPERS SITE SOIL SAMPLES

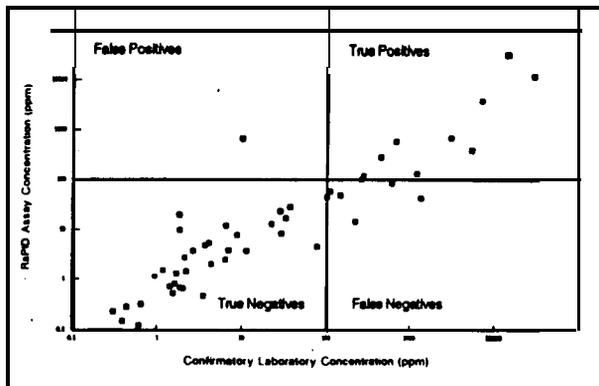
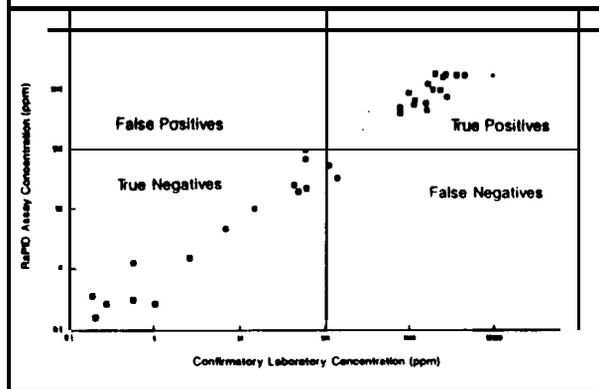


FIGURE 7-3. WINONA POST SITE SOIL SAMPLES



sets. The Wilcoxon Signed Ranks Test was used to verify these results. It indicated that the assay kit's data was significantly different from that of the confirmatory laboratory. These results indicate that this technology is not accurate, but its results can be mathematically corrected to estimate corresponding confirmatory data. Based on these results, 10 to 20 percent of the samples analyzed by this method need confirmation analysis so that the regression parameters can be defined. This places this technology, for the combined soil data set, into the Level 2 data quality category. Figure 7-1 shows the relationship between the assay kit's data and the confirmatory laboratory's data. All data points are

**TABLE 7-14. SUMMARY OF REGRESSION AND RESIDUAL STATISTICS: OHMICRON**

	N	r <sup>2</sup>	Y-int	Slope	Standard Error	Wilcoxon Probability
All Data	84	.81	28	.43	197	Significant Difference
All Data <100 ppm	46	.76	.55	.71	6.1	Significant Difference
All Data >100 ppm	33	.68	71	.43	350	Significant Difference
Koppers-All Data	<b>48</b>	.65	-3.4	.37	350	Significant Difference
Koppers <100 ppm	32	.61	1.5	.57	4.7	Significant Difference
Koppers >100 ppm	13	.60	-110	.39	680	Significant Difference
Winona-All Data	<b>33</b>	.90	34	.51	<b>160</b>	Significant Difference
Winona <100 ppm	15	.69	-3.8	1.2	22	No Significant Difference
Winona >100 ppm	20	.75	<b>98</b>	.46	280	Significant Difference

Notes:

- N** Number of data points.  
**r<sup>2</sup>** Coefficient of determination adjusted for variance.  
**Y-int.** Y-axis intercept of the regression line.  
**Standard Error** Standard error of the estimate.

shown. This figure has been divided at 100 ppm to show the response of the kit relative to a common action level.

The second tier of the evaluation involved the separation of the data by site. This evaluation was conducted to assess potential carrier effects on a technology's performance. Figures 6-2 and 6-3 illustrate these assessments. The initial analysis on the data produced by samples from the former Koppers site was based on results from 50 samples. The r<sup>2</sup> for this regression was 0.48, indicating that a relationship may exist between the data sets. A residual analysis of the data identified samples 47 and 48 as outliers. PRC removed these two points as outliers and recalculated the linear regression. When the regression was recalculated on the 48 remaining sample results, it defined an r<sup>2</sup> factor of 0.65, indicating that some relationship may exist between the two data sets. The Wilcoxon Signed Ranks Test verified these results. It indicated that the kit's data was significantly different from that of the confirmatory laboratory. This data indicates that the technology is not accurate, and places it, for the former Koppers site soil data set, into the Level 1 data quality category.

The initial linear regression analysis on the data produced by samples from the Winona Post site was based on results from 38 samples. The r<sup>2</sup> for this regression was 0.66, indicating that a relationship may exist between the data sets. A residual analysis of the data identified samples 59, 61, 70, 76, and 77 as outliers. PRC removed these 5 points and recalculated the linear regression. When the regression was recalculated on the 33 remaining results, it defined an r<sup>2</sup> factor of

0.90, indicating that this technology meets the first criteria for Level 3 data classification. The slope (0.51) and the y-intercept (34.4) are not both statistically similar to their expected values for Level 3 classification. The Wilcoxon Signed Ranks Test was used to verify these results. It indicated that the kit's data was significantly different from that of the confirmatory laboratory. This indicates that the kit's data must be mathematically corrected to become accurate. This indicates that this technology is not accurate but its results can be mathematically corrected to estimate corresponding confirmatory data. Generally, 10 to 20 percent of the samples analyzed by this technology would need to be submitted for confirmatory analysis to define the regression parameters necessary for the predictive model. This factor places this technology, for the Winona Post soil data set, into the Level 2 data quality category. Based on these results for the data set as a whole and divided by site, there appears to be a carrier effect. The kit showed a stronger correlation to confirmatory data for samples with PCP in a diesel carrier.

Finally, PRC assessed the data by concentration range. The confirmatory laboratory found that 52 samples had concentrations of less than 100 ppm. The initial linear regression on these samples, defined an r<sup>2</sup> of 0.00, indicating that no relationship exists between the two data sets. A residual analysis of the data, though, identified samples 14, 38, 56, 66, 73, and 96 as outliers. PRC removed these six points and recalculated the linear regression. The r<sup>2</sup> improved to 0.76, indicating that a relationship exists between the two data sets. The

Wilcoxon Signed Ranks Test verified these results. It indicated that the assay kit's data was significantly different from that of the confirmatory laboratory. This indicates that the kit's data must be mathematically corrected to simulate confirmatory data. Based on these results, 10 to 20 percent of samples analyzed by this method need confirmation analysis to calculate the regression parameters for the predictive model. This factor places this technology into the Level 2 data quality category for samples with concentrations less than 100 ppm.

The data set of all results greater than 100 ppm PCP contained 38 samples. The initial linear regression defined an  $r^2$  of 0.42, indicating that a relationship may exist between the two data sets. A residual analysis of the data, identified samples 21, 44, 47, 48, and 59 as outliers. PRC removed these five points and recalculated the linear regression. When the regression was recalculated, it defined an  $r^2$  of 0.68, below the 0.75 necessary for Level 2 classification. This indicates that too little of a relationship exists between the two data sets to produce a useable predictive model. The Wilcoxon Signed Ranks Test indicated, at a 90 percent confidence level, that the kit's data was significantly different from that of the confirmatory laboratory. This data indicates that there is a concentration effect for these samples. For samples with concentrations of PCP below 100 ppm the technology produced Level 2 data; for those with concentrations above 100 ppm, the technology produced Level 1 data.

Of the samples from the former Koppers site, the confirmatory laboratory found that 35 had concentrations of less than 100 ppm. The initial linear regression on these samples defined an  $r^2$  of 0.00. A residual analysis of the data, though, identified samples 14, 38, and 56 as outliers. PRC removed these three points as outliers and recalculated the linear regression. An  $r^2$  of 0.61 was then defined, indicating that a weak relationship exists between the two data sets. The Wilcoxon Signed Ranks Test, at a 90 percent confidence level, verified that the test kit's data was significantly different from that of the confirmatory laboratory. This factor places this technology, for the Koppers samples with less than 100 ppm of PCP, into the Level 1 data quality category. Of the samples from the former Koppers site, the confirmatory laboratory found that 15 had results greater than 100 ppm. The initial linear regression on these samples defined an  $r^2$  of 0.39, indicating that a weak relationship exists between the two data sets. A residual analysis of the data identified samples 47 and 48 as outliers. PRC removed these two points, recalculated the linear regression, and defined a new  $r^2$  of 0.60, indicating little improvement in the relationship between the two data sets. The Wilcoxon Signed Ranks Test

verified that the data sets were significantly different. This factor places this technology, for the former Koppers site samples with more than 100 ppm PCP, into the Level 1 data quality category. This data indicates that there is no concentration effect for the samples from the former Koppers site. At both concentration ranges, the technology produced Level 1 data.

The results from the Winona Post site samples were grouped in the same way. The data set for results of less than 100 ppm contained 15 samples, defined an  $r^2$  factor of 0.69, and identified no outliers. However, the Wilcoxon Signed Ranks Test, at a 90 percent confidence level, indicated that the assay kit's data was not significantly different from that of the confirmatory laboratory. The contradiction between the two tests indicates that one or more of the data sets are not distributed normally, violating a fundamental assumption of regression analysis. This makes the regression results suspect. Therefore, based on the Wilcoxon Signed Ranks Test, the technology's data can be placed into a Level 2 category. The confirmatory laboratory found that 22 samples from the Winona Post site had results greater than 100 ppm. The initial linear regression on these results defined an  $r^2$  factor of 0.71, indicating that a relationship may exist between the two data sets. Residual analysis of the data identified samples 59 and 76 as outliers. PRC removed these two points as outliers and recalculated the linear regression. The new regression defined an  $r^2$  of 0.75, just within the Level 2 cutoff criteria. This indicates that a relationship exists between the two data sets, allowing a useable predictive model to be produced. The Wilcoxon Signed Ranks Test was used to verify these results. It indicated that the test kit's data was significantly different from that of the confirmatory laboratory. These results indicate that this technology is not accurate, but its results can be mathematically corrected to estimate corresponding confirmatory data. Therefore, 10 to 20 percent of the samples analyzed by this method need confirmation analysis. This factor places this technology, for the combined soil data set for Winona Post samples with PCP concentrations above 100 ppm, into the Level 2 data quality category. Based on this concentration-related data analysis; concentration levels appeared not to affect the technology on these samples. Level 2 data were produced on samples in both ranges.

### ***Soil Samples: Precision***

Intermethod precision was assessed by a comparison of the assay kit's results on duplicate samples to similar results from the confirmatory laboratory. Three assessments were done; one on all duplicate pairs, and one on the samples from each site. Due to the small sample size for duplicates when grouped by site, those evaluations have less statistical power than the evaluation of the

combined data set. The evaluation of precision on a site-specific basis, though, reveals precision trends and possibly identifies potential PCP carrier influences on precision. When the Dunnett's Test compared the RPDs between all of the field duplicates, it found that the precisions were similar. When precision was examined relative to the site from which the duplicates were collected, the Dunnett's Test again showed that the precisions were similar. This indicates that the technology's precision is not different from the confirmatory laboratory's, for all data groupings. The Wilcoxon Rank Sum Test confirmed this.

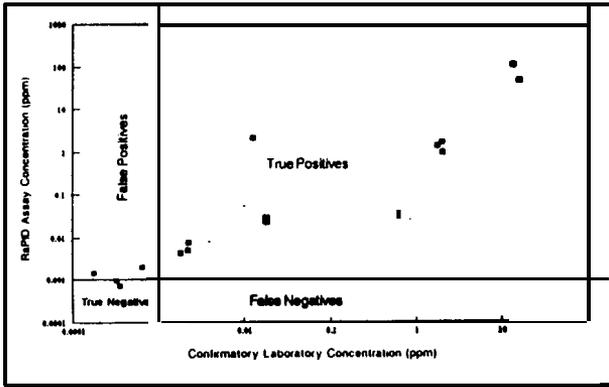
**Water Samples: Accuracy**

This section presents an assessment of accuracy by tier. Tier 1 involves the examination of the data set as a whole. Tier 2 involves the examination of data sorted by site and is intended to give an indication of possible PCP carrier effects on the technology's performance.

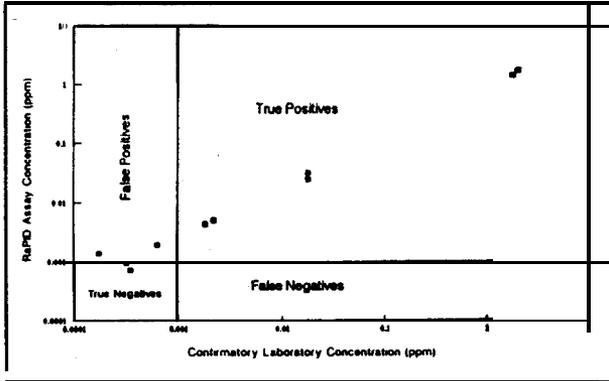
The initial linear regression analysis was based on results from 19 samples. The  $r^2$  for this regression was 0.75, indicating that a relationship exists between the data sets. Figure 7-4 shows this relationship relative to the MCL of 1 ppb. A post-hoc analysis of residuals identified samples 102 and 103 as outliers. The PCP concentrations detected in these samples by the assay test were both more than 10 times greater than their corresponding confirmatory results. These two points were removed and the regression was run again. The second regression analysis produced an  $r^2$  of 0.33, a slope of 0.60, and a y-intercept of 0.14. The reduction in the  $r^2$  value out of the acceptable range for Level 2 data, shows that these two points were significantly biasing regression analysis. However, the Wilcoxon Signed Ranks Test did not verify these results. Both the original data set and the data set with the outliers removed indicated, at a 90 percent confidence level, that the assay hit's data was not significantly different from-that of the confirmatory laboratory. The contradiction between the regression analysis and the Wilcoxon Signed Ranks test indicates that one or both of the data sets do not meet the assumption of normality essential for accurate regression analysis. Therefore, the regression data was considered suspect. Based on the Wilcoxon Signed Ranks Test, this technology, for the combined water data set, falls into the Level 2 data quality category.

Ten samples from the former Koppers site were analyzed. The initial linear regression analysis on these samples resulted in an  $r^2$  of 0.99, indicating that a strong relationship exists between the data sets (Figure 7-5). This  $r^2$  value meets the first condition for a technology to be classified as capable of producing Level 3 analyti-

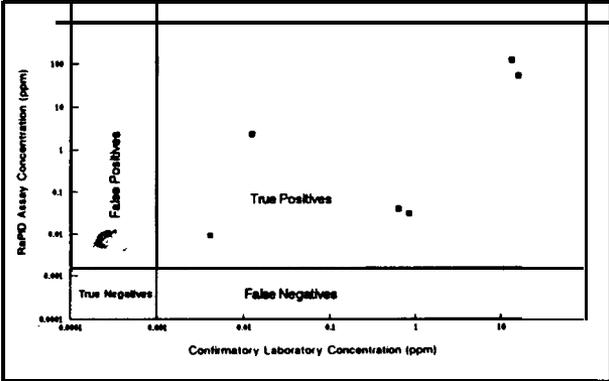
**FIGURE 7-4. TOTAL WATER DATA SET**



**FIGURE 7-5. WINONA POST SITE WATER SAMPLES**



**FIGURE 7-6. WINONA POST SITE WATER SAMPLES**



cal data. However, an examination of the slope (0.86) and y-intercept (0.003) showed that both of these parameters are not statistically equivalent to their expected values for a Level 3 analytical method. The Wilcoxon Signed Ranks Test indicated that the assay kit's data was not significantly different from that of the confirmatory laboratory. All of this indicates that the technology is not accurate, but it does produce Level 2 data. The data produced from this technology can be corrected if 10 to 20 percent of the samples are submitted for confirmatory analysis.

Six samples were from the Winona Post site. The initial linear regression analysis on these samples re-

Six samples were from the Winona Post site. The initial linear regression analysis on these samples resulted in an  $r^2$  of 0.65, indicating that a relationship may exist between the data sets (Figure 7-6). However, this data set contains the two points identified as significant outliers when the combined data set was evaluated. These points also were identified as outliers for this subset. Removal of these points reduced the  $r^2$  to 0.00 indicating that no relationship exists. This reduction in  $r^2$  shows that these two data points strongly biased the initial regression analysis. The  $r^2$  based on the smaller data set confirms that this data does not meet either Level 2 or Level 3 criteria. This confirmed the regression analysis and indicated that the assay kit's data was not accurate for the Winona Post site water sample analysis. This data, therefore, does not meet either Level 2 or Level 3 criteria. This data falls into the Level 1 category.

The water data indicates that a carrier effect may exist. The accuracy of this technology was greater for the samples where isopropyl ether and butane was the PCP carrier solvent.

### ***Water Samples: Precision***

When the Dunnett's Test compared the RPDs between the technology's entire field duplicate data set and the corresponding confirmatory laboratory data set, the data sets were found to be statistically similar. This indicates that the technology's precision is not different from the confirmatory laboratory's. The Wilcoxon Signed Ranks Test confirmed this data. The data set was not reassessed by site because only one duplicate pair was collected from the Winona Post site.

## Section 8

### Millipore Corporation: EnviroGard PCP Test Kit

The EnviroGard PCP Test Kit is designed to quickly provide semiquantitative or quantitative results for PCP concentrations in soil and water samples. In its standard configuration for soil analysis, the kit detects PCP concentrations in the following ranges: greater than 5 ppm, between 5 and 0.25 ppm, between 0.25 and 0.025 ppm, and less than 0.025 ppm. For water analysis, the kit detects PCP in the following ranges: greater than 100 ppb, between 100 and 20 ppb, between 20 and 5 ppb, and less than 5 ppb. The developer will customize these ranges to meet specific needs.

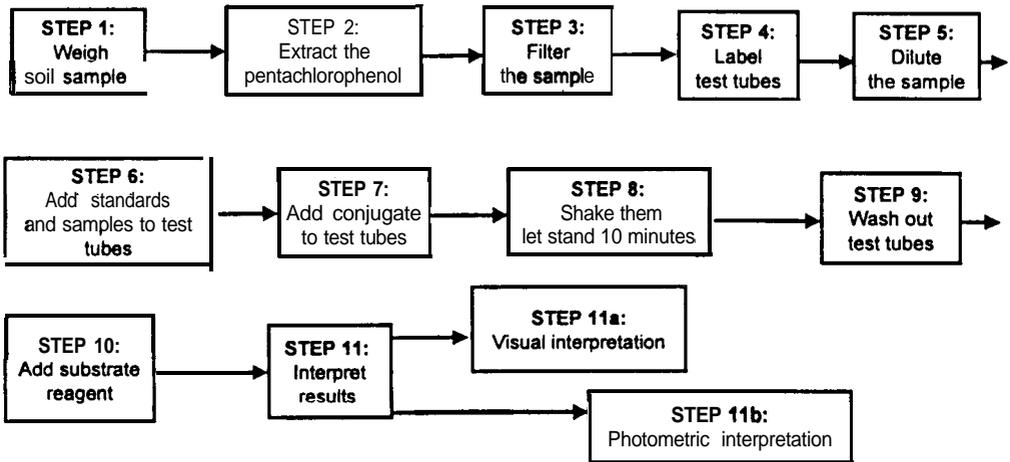
The kit uses the principles of ELISA to determine PCP concentrations in water and soil samples. A review of the principles of ELISA is presented in Section 5. Unlike the EnSys technology, though, the colors produced by samples tested by the kit are compared to three PCP standards taken through all of the immunoassay steps (Exhibit 8-1). The results can be determined visually or by using a differential photometer. For quantitative comparisons, the sample and standards are compared to a blank water sample using the differential photometer. The differential photometer readings are then be graphed as a standard curve, allowing for the quantitative determination. Overall, the kit is portable and can be operated outdoors, but temperature extremes and humidity can affect their performance. The reagents used for sample analysis require refrigeration. The differential photometer requires electricity but can be operated using a rechargeable battery. The kit was found to be easy to operate even by individuals with no prior PCP immunoassay testing experience.

The highest number of demonstration samples analyzed in one 10-hour day was 43. The average number of demonstration samples analyzed in one 10-hour day was 20. Sample throughput was higher when using the kit to obtain semiquantitative results. Quantitative results require dilutions and reanalyses of samples which decreases sample throughput. The detection limit reported by the developer for soil samples is 0.025 ppm and for water samples is 5 ppb.

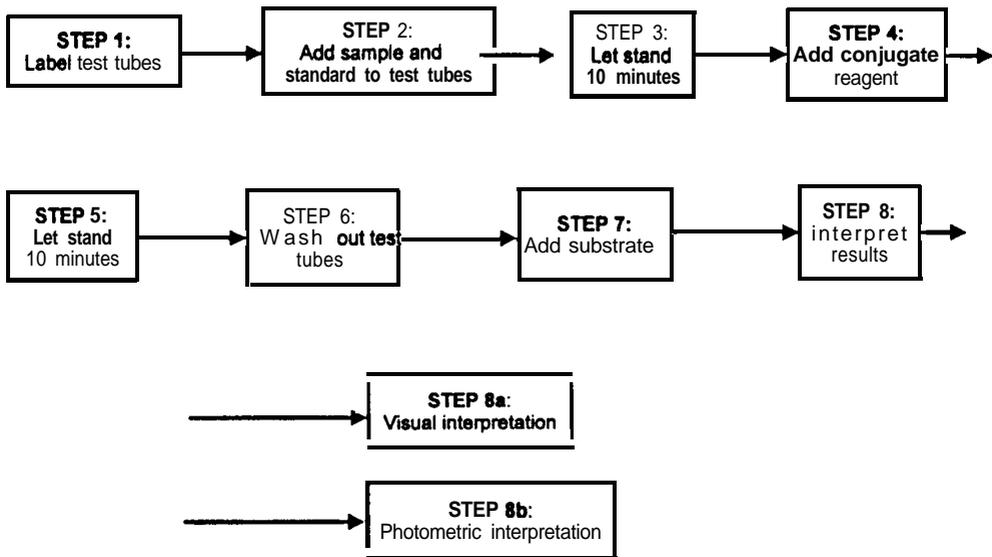
The intramethod and intermethod statistical comparisons for this technology revealed that it produced a high percentage of false negative results and that they had difficulty reproducing their results. A draft version of these results was issued on March 4, 1994. The developer submitted comments on the draft version on May 17, 1994. In its comments, Millipore stated: "Upon reviewing the data we were extremely concerned with the lack of agreement with the reference method particularly in regard to the number of false negative results. We immediately began an investigation into the source of these discrepancies." The developer noted that the concentrations encountered during the demonstration, especially those above 1,000 ppm, "significantly exceeded concentrations we used in fortified samples used to develop the immunoassay when there is more emphasis on assay sensitivity." The developer then fortified samples to these concentrations in the laboratory and "noted two phenomena: there was a high incidence of false negative results and there was excessive variation between duplicate determinations." The developer theorized that this was due to hydrophobic binding of the enzyme conjugate. As a solution, the developer proposed replacing a deionized water wash with a wash that uses a detergent and adding an additional dilution step to the technology's protocols. The developer claims these changes greatly improved the technology's laboratory results. The developer then used the new protocol on about 33 extracts left over from the demonstration. "Using the revised protocol, the false negative rate was reduced to 3 percent while the false positive rate remained at 3 percent." The developer stated that the new protocol has now been included in its commercial product. It added, "We believe that, based on the knowledge gained from the field demonstration, we have been able to make significant improvements in our product. This experience demonstrates the utility of the SITE program and its goal of evaluating innovative technologies." It should be noted that the SITE Program has not yet reevaluated the EnviroGard PCP Test Kit since the change in protocol.

Exhibit 8-1. The processes for the Millipore technology.

### MILLIPORE ENVIROGARD PENTACHLOROPHENOL SOIL TEST KIT PROCEDURES



### MILLIPORE ENVIROGARD PENTACHLOROPHENOL WATER TEST KIT PROCEDURES



## Section 8

### Applications Assessment

The principal advantage of the ELISA technology is that it is very specific to PCP. This specificity reduces the chances of determining that a sample contains PCP, when in fact it does not. This specificity also greatly reduces the chances of determining that a sample contains no PCP, when it in fact does. ELISA technologies are generally inexpensive when compared to formal laboratory analysis using EPA-approved methods for PCP. They are generally simple to operate even for individuals with no prior immunoassay testing experience. These technologies are portable and can be operated outdoors under certain conditions. By batching samples ELISA methods can have high sample throughput, and they are capable of quickly providing sample results.

ELISA systems are most applicable to sites where PCP is a known contaminant and where large concentrations of other chemicals are not present in the samples. Generally, the larger the number of samples to be collected, the greater the advantage of using these systems. The use of these systems at these sites can often decrease the cost of an investigation by decreasing the number of samples requiring confirmatory laboratory analysis and by enabling more work to be completed during the sampling visit. Using these systems can allow work to continue without having to wait for confirmatory laboratory results. These systems also can be used by laboratories as screening tools for PCP. Results can be used to determine appropriate sample extraction techniques, as well as to determine dilutions which may be required for sample analysis. These results also can be used to determine the appropriate analytical method to be used for sample analysis.

Both semiquantitative and quantitative ELISA systems are available. The semiquantitative systems provide sample results greater than or less than a specific detection level, while the quantitative systems provide estimates of actual contaminant concentrations. The detection levels for the systems can be customized to meet site-specific action levels. For quantitative systems, more than one detection level can be used to obtain

a closer estimate of PCP concentrations in samples. A potential limitation of ELISA systems is that their results may not always agree with results from the analysis of the same sample by EPA-approved methodologies. These systems' results, when compared to confirmatory laboratory results, included both false positive results, results which overestimate the concentration of PCP in the sample, and false negative results, results which underestimate the concentration of PCP in the sample. Both false positive and false negative results have important implications on investigative and remedial activities. Another general limitation of ELISA systems is that some organic compounds, particularly tetrachlorophenols and trichlorophenols, will provide a positive response when present in a sample at part per million levels.

These systems also can be affected by other chemicals found naturally in environmental samples, such as humic acids, and by chemicals which are associated with PCP treatment of wood products, such as petroleum hydrocarbon products and solvents. These chemicals must generally be present in percent level concentrations before these systems are affected. Logistical limitations of these systems generally include that the enzyme conjugate used for soil and water sample analysis requires refrigeration. The ELISA technologies are generally temperature sensitive and should not be operated in temperature extremes. Temperatures less than 32 °F will cause certain reagents to freeze, and temperatures greater than 97 °F may cause inaccurate results. High humidity also may affect the immunoassay chemistry.

#### **EnSys Inc.:** **Penta RISc Test System**

**This** system is designed to provide semiquantitative screening results for PCP in water and soil samples. This system's soil test kit does not require refrigeration. This is a specific advantage of this technology. During the demonstration, this system appeared to be highly temperature sensitive. When ambient temperatures

approached 90 °F, the system's calibration failed at a high **frequency**. Therefore, during the summer, the use of this technology in an air-conditioned environment is advised.

For the soil sample analysis, this system produced its highest percentage of correct readings for samples contaminated with PCP in an isopropyl ether and butane carrier solvent. However, this carrier also produced a higher percentage of false negatives with this carrier relative to the diesel carrier. For the water sample analysis, this system produced a higher percentage of correct readings and a lower percentage (0 percent) of false negative results for samples contaminated with PCP in an isopropyl ether and butane carrier relative to a diesel carrier.

Overall, the results of the demonstration indicate that the system does not meet its developer's performance specifications and its data is statistically different from corresponding confirmatory data. Based on this, the system produces Level 1 screening data. This classification is based on the technology's failure to meet its developer's performance specifications. This demonstration indicated that for soils, the system tended to give false negative results when used on samples with PCP concentrations below 10 ppm. All of the remaining inaccuracies were false positive results. Based on these results, if target action levels are greater than 10 ppm PCP, and when false positive results are acceptable, the system can be used to guide field work and sampling efforts. These activities include: determining the vertical and horizontal extent of PCP contamination in soil, tracking PCP groundwater contamination plumes, and determining PCP contamination in surface waters. Another use of the system is to monitor the effectiveness of remediation techniques employed to reduce or eliminate PCP contamination. The Penta RISC Test System can be used to determine whether PCP concentrations in soil or water samples exceed site-specific action limits. In such applications, the results would be considered conservative based on the potential frequency of false positive results. False positive results will cause remediation efforts to be performed in areas that do not require them. This is considered a conservative error compared to false negative results which cause no remediation to take place in areas where it is needed. Field investigators must realize that the system is designed as a screening tool to assist in evaluating PCP contamination. It is an approved method for determining whether PCP concentrations are above or below a site-specific action limit. Limited confirmatory laboratory analysis, using EPA-approved methods, will provide QC and check the performance of the technology.

## **Ohmicron Corporation: Penta RaPID Assay**

The PCP RaPID Assay is designed to provide quantitative screening results for PCP in water and soil samples. Applications for the assay include both laboratory and field uses. During the demonstration, this system appeared to be temperature sensitive. Therefore, during summer, the use of the assay in an air-conditioned environment is advised.

This demonstration indicated that this technology's soil results may be affected by the type of carrier used. When the carrier solvent was diesel fuel, this technology produced Level 2 data. This means that its data can be used to guide field work and sampling efforts. These activities include determining the vertical and horizontal extent of PCP contamination in soil, tracking PCP groundwater contamination plumes, and determining PCP contamination in surface waters. Another use of the system is to monitor the effectiveness of remediation techniques employed to reduce or eliminate PCP contamination. The PCP RaPID Assay can be used to determine whether PCP concentrations in soil or water samples exceed site-specific action limits. In such applications, the results would be considered conservative based on the potential frequency of false positive results, unless the data was corrected through confirmatory analysis. False positive results will cause remediation efforts to be performed in areas that do not require them. This is considered a conservative error compared to false negative results which cause no remediation to take place in areas where it is needed. Field investigators must realize that the system is designed as a screening tool. It is a field method for determining PCP concentrations. Limited confirmatory laboratory analysis, using EPA-approved methods, will allow data correction, provide QC, and check the performance of the technology.

For samples where isopropyl ether and butane were the PCP carrier solvent, the assay produced Level 1 data. In this application, the technology only could be used to detect the presence or absence of PCP. Therefore, its field uses would be limited to initial site survey activities only. The results and carrier effects identified through the soil analysis were reversed for the water analysis.

## **Millipore Corporation: EnviroGard PCP Test Kit**

This test kit can be used on both soil and water samples. It was developed as a semiquantitative field

test, but was assessed during this demonstration for its ability to produce both semiquantitative and quantitative results. During the demonstration, this test was found to produce false negative results and poor precision when high concentrations of PCP were found in a sample. As a result, the kit's developer has changed part of the

analytical protocol, including adding a wash step that uses a detergent. The developer claims that the modified EnviroGard PCP Test Kit has only a 3 percent false negative rate and a 3 percent false positive step, but the SITE Program has not yet reevaluated the kit since the modification.

## Section 9 References

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