THE ENVIRONMENTAL TECHNOLOGY VERIFICATION PROGRAM

U.S. Environmental Protection Agency

The Business of Innovation

ETV Joint Verification Statement

TECHNOLOGY TYPE:	**PORTABLE CYANIDE ANALYZER**
APPLICATION:	**DETECTING CYANIDE IN WATER**
TECHNOLOGY NAME:	**Cyanide ReagentStrip™ Test Kit**
COMPANY:	**Industrial Test Systems, Inc.**
ADDRESS:	**1875 Langston Street Rock Hill, SC 29730**
PHONE:	**803-329-9712**
FAX:	**803-329-9743**
WEB SITE:	**http://www.sensafe.com**
E-MAIL:	**its@cetlink.net**

The U.S. Environmental Protection Agency (EPA) supports the Environmental Technology Verification (ETV) Program to facilitate the deployment of innovative or improved environmental technologies through performance verification and dissemination of information. The goal of the ETV Program is to further environmental protection by substantially accelerating the acceptance and use of improved and cost-effective technologies. ETV seeks to achieve this goal by providing high-quality, peer-reviewed data on technology performance to those involved in the design, distribution, financing, permitting, purchase, and use of environmental technologies.

ETV works in partnership with recognized standards and testing organizations, with stakeholder groups (consisting of buyers, vendor organizations, and permitters), and with individual technology developers. The program evaluates the performance of innovative technologies by developing test plans that are responsive to the needs of stakeholders, conducting field or laboratory tests (as appropriate), collecting and analyzing data, and preparing peer-reviewed reports. All evaluations are conducted in accordance with rigorous quality assurance (QA) protocols to ensure that data of known and adequate quality are generated and that the results are defensible.

The Advanced Monitoring Systems (AMS) Center, one of six technology areas under ETV, is operated by Battelle in cooperation with EPA's National Exposure Research Laboratory. The AMS Center has recently evaluated the performance of cyanide analyzers used to detect cyanide in water. This verification statement provides a summary of the test results for the Industrial Test Systems, Inc., Cyanide ReagentStrip™ test kit.

VERIFICATION TEST DESCRIPTION

The verification was based on comparing the cyanide concentrations of water samples determined by the Cyanide ReagentStrip™ test kit with cyanide concentrations determined by a laboratory-based reference method (EPA Method 335.1, *Cyanides Amenable to Chlorination*). The verification test was conducted between September 22, 2004, and October 5, 2004; and the results were compared to assess accuracy, precision, matrix effects, linearity, method detection limit (MDL), inter-unit reproducibility, lethal or near-lethal dose response, operator bias, field portability, ease of use, and sample throughput. Samples used in the verification test included quality control samples, performance test (PT) samples [ranging in concentration from 0.03 to 25 milligrams per liter (mg/L)], lethal/near-lethal concentration samples (at concentrations of 50, 100, and 250 mg/L), drinking water (DW) samples, and surface water samples. The results from the Cyanide ReagentStrip™ test kit were compared with the reference method to quantitatively assess accuracy and linearity. Multiple aliquots of each test sample were analyzed separately to assess the precision of both the Cyanide ReagentStrip™ test kit and the reference method. Seven non-consecutive replicate analyses of a 0.05-mg/L solution were made to obtain precision data with which to determine the MDL. Two Cyanide ReagentStrip™ test kits were tested independently, and the results were compared to assess inter-unit reproducibility. The Cyanide ReagentStrip™ test kit was tested by a technical and a non-technical operator to assess operator bias. Sample throughput was estimated based on the time required to analyze a sample. Ease of use was based on documented observations by the operators and the Battelle Verification Test Coordinator. The Cyanide ReagentStrip™ test kit was used in a field environment, as well as in a laboratory setting, to assess the impact of field conditions on performance.

QA oversight of verification testing was provided by Battelle. Battelle QA staff conducted a technical systems audit, a performance evaluation audit, and a data quality audit of 10% of the test data.

TECHNOLOGY DESCRIPTION

The following description of the Cyanide ReagentStrip™ test kit was provided by the vendor and does not represent verified information.

The Cyanide ReagentStrip™ test kit is designed to detect free cyanide in water. This is done by converting cyanide in water to cyanogen chloride, which, in the presence of isonicotinic and barbituric acids, produces a color change that can be detected visually or with a colorimeter. Results can be determined by three methods, and the method selected is dependent upon the data needs of the user: (1) A semi-quantitative result in increments ranging from <0.1 mg/L to >10 mg/L is obtained in approximately 1 minute by comparing the color change on ReagentStrip™ #2 to a color chart; (2) a semi-quantitative result for an expanded range of 0 mg/L to >200 mg/L may also be obtained in 10 minutes by visually comparing the color of the water sample in a microcuvette with a separate color chart designed for use with microcuvettes; and (3) a quantitative determination can be obtained in 10 minutes when the microcuvette is inserted into the optional ReagentStrip™ CO7500 colorimeter (also identified as the ReagentStrip™ Reader), and the intensity of the color is measured quantitatively. The ReagentStrip™ Reader generates a result in absorbance units that are converted to concentration units using the reference table provided by Industrial Test Systems, Inc. The absorbance units on the reference table convert to concentrations ranging from <0.01 mg/L to >60 mg/L. The kit includes one bottle each of Cyanide ReagentStrip™ #1 and #2, one graduated pipette, 20 microcuvettes, one microcuvette holder, one ReagentStrip™ Reader, two semi-quantitative visual color charts, one ReagentStrip™ Reader absorbance reference chart, one instruction sheet, and one material safety data sheet. The list price, including the optional ReagentStrip™ Reader, is $559.99 for 50 tests. ReagentStrip™ #1 and #2 for additional tests can be purchased separately at an approximate cost of $40 for 50 additional tests.

VERIFICATION OF PERFORMANCE

Accuracy: The quantitative accuracy of the ReagentStrip™ Reader-derived results were evaluated by calculating biases with respect to the reference concentrations. Biases for the Cyanide ReagentStrip™ test kit ranged from -47 to 25% for the PT samples; -27 to 28% for the surface water samples; -41 to 3% for the DW samples from around the United States; and, with the exception of the samples analyzed outdoors, -42 to 30% for the Columbus, OH, DW samples. The matrix effect on these results was apparently minimal because the range of the bias for the surface water and DW results was similar to that of the PT samples that were prepared in deionized water. For the semi-quantitative accuracy results, 84% of the PT sample results matched the exact color that should represent the reference concentration, and 16% were within one color on the color chart; 100% of the surface water and U.S. DW samples matched the exact color; and 83% of the Columbus, OH, DW samples matched exactly, with the remaining 17% within one color. For the microcuvette results, 64% of the PT samples matched exactly and 36%

were within one color; 56% of the surface water samples matched exactly, with 44% being within one color; 88% of the U.S. DW samples matched exactly, and 12% were within one color; and 66% of the Columbus, OH, DW samples matched exactly, 17% were within one color, and 17% were within two colors on the color chart.

Precision: The quantitative precision of the Cyanide ReagentStrip™ test kit using a ReagentStrip™ Reader was evaluated by calculating relative standard deviations (RSDs) for each sample set. The RSDs ranged from 4 to 86% for the PT samples (if the 0.03 mg/L samples are removed, the upper end of that range was 27%); 11 to 30% for the surface water samples; 3 to 25% for the DW samples from around the United States; and, except for the outdoor field site, 2 to 21% for the Columbus, OH, DW samples. The RSDs were similar regardless of the water matrix. For the semi-quantitative strip evaluation, the precision was evaluated by determining the frequency by which the qualitative result (color) was produced for each sample set. The same result was obtained for four replicates in 11 out of 16 PT sample sets, all of the surface water sample sets, all 10 U.S. DW sample sets, and 11 out of 12 sample sets of Columbus, OH, DW. For the microcuvettes, consistent results were obtained for 11 of 16 PT sample sets, 2 out of 4 surface water sample sets, 9 out of 10 U.S. DW sample sets, and 7 out of 12 sample sets of Columbus, OH, DW.

Linearity: A linear regression of the PT sample results, ranging in concentration from 0.03 to 25 mg/L, gave the following regression equations:

y (non-technical operator results in mg/L) = 1.03 (± 0.06)x (reference result in mg/L) + 0.19 (± 0.55) mg/L with r^2 = 0.947 and N = 72.

y (technical operator results in mg/L) = 0.97 (± 0.04)x (reference result in mg/L) + 0.08 (± 0.42) mg/L with r^2 = 0.965 and N = 72.

where the values in parentheses represent the 95% confidence interval (two times the standard error) of the slope and intercept, r^2 is the coefficient of determination, and N is the total number of results with corresponding reference analyses. The slopes of these regressions are not significantly different from unity, neither intercept is significantly different from zero, and the r^2 values are both above 0.94. Linear regressions were also generated for a concentration range of 0.03 to 1 mg/L. Within this smaller concentration range, both slopes were significantly different from unity, while the intercepts were either not significantly different from zero or very close to it. This suggests that the non-technical operator's results have a slightly high bias, and the technical operator's results have a slightly low bias. This effect was not apparent with the wider concentration range.

Method Detection Limit: The quantitative MDL of the Cyanide ReagentStrip™ test kit was determined by analyzing seven replicate samples at a concentration of 0.05 mg/L. The MDLs were 0.04 and 0.03 mg/L for the non-technical operator and 0.02 for both units when used by the technical operator. The strips generated detectable results at all concentration levels of 0.2 mg/L and above, and the microcuvettes generated detectable results at all concentrations of 0.03 mg/L and above.

Inter-Unit Reproducibility: A linear regression of the data for the inter-unit reproducibility assessment gave the following regression equation:

y (Unit #1 result in mg/L) = 1.17 (± 0.02)x (Unit #2 in mg/L) - 0.08 (± 0.08) mg/L with r^2 = 0.991 and N = 213.

The intercept was not significantly different from zero; however, the slope was significantly larger than unity, suggesting that, on average, Unit #1 gave slightly higher responses than Unit #2.

Lethal/Near-Lethal Dose Response: The ReagentStrip™ Reader results for lethal to near-lethal concentrations ranged from 36 to >60 mg/L. According to the vendor, concentrations greater than 40 mg/L exceed the linear range of the ReagentStrip™ Reader so these results would need to be clarified through dilution and reanalysis of samples. For the semi-quantitative strips, all 12 samples for both operators correctly resulted in a >10-mg/L (the highest concentration color) result. For the microcuvettes, all 12 samples tested by the technical operator resulted in >20-mg/L results, but the 250-mg/L samples did not appear to change to the >200-mg/L color. If the sample analysis procedure was only observed visually, the color changes on the strips and of the solution (for the lethal dose concentration) in the microcuvettes were quick and pronounced with respect to the much lower concentrations. In this manner, the presence of a high concentration of cyanide was easily ascertained through use of all three detection mechanisms.

Operator Bias: A linear regression of the data for the operator bias assessment gave the following regression equation:

> y (non-technical operator result in mg/L) = 1.04 (± 0.04)x (technical operator result in mg/L) + 0.08 (± 0.26) mg/L with r^2 = 0.936 and N = 167.

This regression, with a slope and intercept that are not different from unity and zero, respectively, suggests that the results generated by both operators were not different from one another. This is consistent with the wider concentration range linearity results. However, the linearity results over the concentration range of 0.03 to 1 mg/L suggested that the non-technical operator's results were biased slightly high, while the technical operator's results were biased slightly low. In this instance, because neither operator's results are necessarily closer to the reference method result (as the linearity data show), it is unlikely that the difference between the two operators was related to differences in training or experience. It is apparently the result of the normal variability of two different people performing the analyses at these lower concentrations. For the semi-quantitative microcuvette results, in only one out of 108 results was one operator's result more than one color different from the other operator's result. For the strips, the results from both operators were always within one color of one another.

Field Portability: No functional aspects of the Cyanide ReagentStrip™ test kit were compromised by performing the analyses in the field setting. However, performing analyses when water temperatures were approximately 17°C negatively affected the performance of the reagents as was evidenced by the large negative biases for the samples analyzed outdoors.

Ease of Use: The Cyanide ReagentStrip™ test kit was easy to operate. The written instructions provided were clear, and the accompanying instructional video (lasting less than 5 minutes) was detailed and easy to understand. Because the required reagents were transferred into the test sample entirely by the repeated dipping of the two types of ReagentStrips™, there was no measuring or mixing. The strips, microcuvettes, and ReagentStrip™ Reader were easy to use; and cleanup was minimal.

Sample Throughput: The analysis of a set of approximately 10 samples, including sample preparation and reaction time, took 30 to 40 minutes.

Original signed by Gabor J. Kovacs 4/15/05
Gabor J. Kovacs Date
Vice President
Energy and Environment Division
Battelle

Original signed by Andrew P. Avel 5/16/05
Andrew P. Avel Date
Acting Director
National Homeland Security Research Center
U.S. Environmental Protection Agency