ENVIRONMENTAL TECHNOLOGY VERIFICATION









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ETV Joint Verification Statement

TECHNOLOGY TYPE: Handheld luminometer

APPLICATION: Toxicity testing of effluent wastewater.

PRODUCT NAME: ECLOX Handheld Luminometer with LUMIStherm

Thermoblock and LUMISsoft4 Software

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Testing and verification of a handheld luminometer for detecting toxicity of wastewater effluent was conducted as a joint verification project with the Danish Centre for Verification of Climate and Environmental Technologies (DANETV), ETV Canada, and the United States Environmental Protection Agency (U.S. EPA) Environmental Technology Verification Program (ETV). The testing and verification satisfied the requirements of the Canadian ETV program, U.S. ETV program and the ETV scheme currently being established by the European Union (EU ETV).

Environment Canada established the Canadian ETV program to provide credible information for promoting the commercialization and market deployment of new environmental technologies, thus helping to address environmental challenges efficiently, effectively and economically. Information on the Canadian ETV program is available at www.etvcanada.ca.

The U.S. ETV program was established to facilitate the deployment of innovative or improved environmental technologies through performance verification and dissemination of information. The goal of U.S. ETV is to further environmental protection by accelerating the acceptance and use of improved and cost-effective technologies. U.S. 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. Information and U.S. ETV documents are available at www.epa.gov/etv.

DANETV was established by four independent Danish research and technology organizations and is supported by the Danish Agency for Science, Technology and Innovation under the Danish Ministry of Science, Technology and Innovation to provide environmental technology verification for vendors of innovative water technologies. Information and DANETV documents are available at www.etv-denmark.com.

DHI as DANETV Water Center performed the testing and verification in collaboration with ETV Canada and the ETV Advanced Monitoring Systems (AMS) Center, managed by Battelle through a cooperative agreement with the U.S. EPA.

VERIFICATION AND TEST DESCRIPTION

Testing and verification of the ECLOX Handheld Luminometer took place between January and April 2010. DHI personnel, with support from the vendor, HACH-LANGE, coordinated and supervised the testing and verification procedure. The testing took place in the DHI laboratories, Hørsholm, Denmark. DHI operated the luminometer during the testing and verification procedure. HACH-LANGE provided the product (luminometer, thermal block and software), user manuals, and operating instructions for the tests. The Eclox verification test was performed simultaneously with the verification test of LUMIStox 300 Bench Top Luminometer also from HACH-LANGE. ETV Canada, Battelle and DHI jointly produced the verification protocol, test plan, process document and report documents including this Verification Statement with input from HACH-LANGE and an independent expert group.

The test program was designed to comply with ISO standard 11348-3: "Water quality – Determination of the inhibitory effect of water samples on the light emission of *Vibrio fischeri* (Luminescent bacteria test)" as well as the instrument manuals. However, the ECLOX with LUMIStherm Thermal block and LUMISsoft4 Software does not fulfill all ISO 11348-3 requirements.

Stock solutions were prepared in 2% NaCl MilliQ water. Solid NaCl was added to wastewater to obtain the salt concentration required for testing with the saltwater bacteria, *Vibrio fischeri*. Dilution series were prepared with dilute saltwater (2% NaCl).

Ten test series in total were performed as outlined in the joint test plan, with deviations mentioned in the report. Each test series provided information on specified performance parameters, as shown in Table 1. One test series was performed on wastewater to provide performance information on wastewater toxicity.

Table 1 Test design and associated performance parameters.

	Matrix		
Performance parameters	2% NaCl MilliQ	Wastewater	
Range, Repeatability, Agreement with accepted values	Χ		
Criterion of detection	Χ		
Robustness, effect of start concentration on repeatability	Χ		
Reproducibility	Χ		
Robustness, sample temperature at field use*	X		
Robustness, sample temperature at laboratory use	Χ		
Robustness, pH	Χ		
Robustness, color	X		
Robustness, turbidity	Х		
Robustness, matrix		Х	

^{*} For this test the ECLOX was not connected to LUMIStherm Thermoblock and LUMISsoft4 Software. ECLOX firmware was used instead of the software. This test was not in compliance with ISO 11348-3.

The operational conditions during testing are shown in Table 2.

Table 2 Operational parameters evaluated during testing.

Temperature of thermoblock	pH in sample	Color correction	Temperature at field use	Wastewater types
14.0 - 16.1°C	6.0 - 8.5	Colored samples	5 - 23°C	Domestic
		Turbid samples		Industrial

Complete information on wastewater quality parameters is included in the verification report.

Quality Assurance

DHI provided internal review of documents and an audit of test performance. Battelle and ETV Canada ensured that the verification and tests were planned and conducted to satisfy the requirements of U.S. ETV and ETV Canada, including input and concurrence from their stakeholder groups. Battelle, U.S. EPA, and ETV Canada quality assurance staff conducted a data quality audit of at least 10% of the test data. Battelle conducted an on-site technical systems audit. Three technical experts provided an independent review of the test plan, and four technical experts reviewed the verification report. This statement reflects the verification results after performing the above quality assurance actions.

TECHNOLOGY AND PRODUCT DESCRIPTION



The following description of the ECLOX Handheld Luminometer is based on information provided by the vendor and does not represent verified information.

The ECLOX is a portable instrument designed to provide data appropriate for risk assessments in the event of environmental releases, emergency situations, preventive security measures, and regulatory monitoring.

The ECLOX is designed in particular to be used for the Luminescent Bacteria Toxicity Test and to be used with a Chemiluminescence Toxicity Test. Both tests will provide results in a short-term and

simple manner in the field or in the laboratory. The ECLOX used in the field gives values of percent inhibition.

The ECLOX can be used to deliver luminescence values by connecting the thermoblock LUMIStherm to a computer program (LUMISsoft4). This approach is used for testing and recording data according to ISO 11348-3 for calculation of the Lowest Ineffective Dilution causing less than 20% inhibition (LID) and Effective Concentration causing 50% inhibition (EC₅₀). In this verification results have been obtained as EC_{50} values or directly as percentage of inhibition.

VERIFICATION RESULTS

Table 3 Description of matrix and effect for ECLOX.

Matrix	Effect
ECLOX is applied for wastewater; river and lake water; leachate from soils and waste; or directly in effluent chemicals. Verification testing was conducted on domestic and industrial wastewater effluents.	Measurement of toxicity as indicated by inhibition of luminescent bacteria by a variety of compounds including metal ions, organic pesticides, inorganic and organic pollutants, and surfactants.
	Additional parameters included: User manual quality, product cost, environmental health and safety.

Performance Parameters

The performance parameters listed in Table 4 are results for EC_{50} values or percent inhibition generated after 30 minutes. Only 30-minute results are included here since they represent the standard length of time for reporting EC_{50} results.

Table 4 ECLOX performance parameter summary.

ECLOX	Criterion of detection	Range of application	Precision		Agreement with accepted values	Robustness		
Compound	% inhibition	mg/L	Repeat- ability %	Reproduc- ibility %	%	pH, color, turbidity, laboratory temperature ¹⁾	Field temperature (15 min)	Waste- water matrix ¹⁾ %
General	5.5							
Zn ²⁺		8.2 - 130	4.9	51	186		11 -171	22 -125
Cr ₂ O ₇ ²⁻		37 - 590	24		96			(-20) -13
Triclosan		1.1 - 17	2.2		190			101 -141
Cyanide		35 - 570	16					
SDS		2.0 - 32	38			85 -115	73 -105	35 -101
CTAB		1.9 -31	1.2		99			61 -73

¹⁾For colored samples, robustness results are presented after the use of color correction. For the BaSO₄-turbid samples, robustness results are presented <u>without</u> the use of color correction. For domestic wastewater, adjustment was made to account for the negative inhibition from the wastewater; if color correction was used the robustness was 101% to 130%. The values listed in table are the best achievable robustness.

General observations included the following:

- The user manual and other instructions were found to be complete.
- The results showed that the use of color correction is essential when testing colored samples, while the results for turbid BaSO₄ samples showed that the color correction function is not applicable. The applicability of color correction for other types of turbid samples was not tested.
- No additional risks for occupational health and environment were noted compared to conventional effluent wastewater testing or analysis.
- The purchase price for the ECLOX, LUMIStherm, the LUMIS of the software at the time of testing was 6,500 Euro (9,600 \$U.S.). Additional equipment such as cuvettes, bacteria and chemicals on a cost-per-sample basis as used for testing for EC₅₀ according to the ISO 11348-3 was 18 Euro (23 \$U.S.).
- The product costs for one laboratory day are listed in Table 5. Shelf life of the dried bacteria is one year, and the lifetime of the rehydrated bacteria is four hours, making it possible to perform an EC₅₀-test according to the ISO 11348-3 on three samples plus associated controls and standards.

Table 5 List of capital cost items and operation and maintenance cost items per product unit.

Item type	Number/duration and item
Capital	
Buildings and land	1 laboratory facility
Equipment	1 ECLOX, 1 LUMIStherm, 1 PC with LUMISsoft4
Utility connections	3 power supplies
Installation	Can be done by operator/laboratory technician on 1 day
Start up/training	Training of laboratory technician, done in 1 day
Operation and maintenance	
Materials, including chemicals	1 bacterial batch, 20 cuvettes per sample (=60), 1 bottle of reconstitution solution, 1 bottle of 2% NaCl solution, 1 bottle of solid NaCl
Utilities, including water and	Power (PC and screen ~6 kWh, ECLOX 4 AA batteries)
energy	
Labor	One laboratory technician for 1 day

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