

## ENVIRONMENTAL TECHNOLOGY VERIFICATION



### ETV Joint Verification Statement

<b>TECHNOLOGY TYPE:</b>	Bench top luminometer	
<b>APPLICATION:</b>	Toxicity testing of wastewater effluent.	
<b>PRODUCT NAME:</b>	LUMIStox 300 Bench Top Luminometer with LUMIStherm Thermoblock and LUMISsoft4 Software	
<b>COMPANY:</b>	HACH-LANGE GmbH	
<b>ADDRESS:</b>	Willstätterstrasse 11 D-40549 Düsseldorf, Germany	<b>PHONE:</b> +49 211 5288 0
<b>WEB SITE:</b>	<a href="http://www.hach-lange.de">www.hach-lange.de</a>	
<b>E-MAIL:</b>	<a href="mailto:elmar.grabert@hach-lange.de">elmar.grabert@hach-lange.de</a>	

Testing and verification of a bench top 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](http://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](http://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](http://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 LUMISTox 300 Bench Top Luminometer took place between January and April 2010. DHI personnel, with support from the vendor, HACH-LANGE, coordinated and supervised testing. The testing took place in the DHI laboratories, Hørsholm, Denmark. DHI operated the luminometer during testing. HACH-LANGE provided the product (luminometer, thermal block and software), user manuals, and operating instructions for the tests. The LUMISTox 300 Bench Top Luminometer verification test was performed simultaneously with the verification test of the Eclox 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.

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.

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

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	Cuvette material	Wastewater types
14.0 - 16.1°C	6.0 - 8.5	Colored samples Turbid samples	Glass Plastic	Domestic Industrial

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

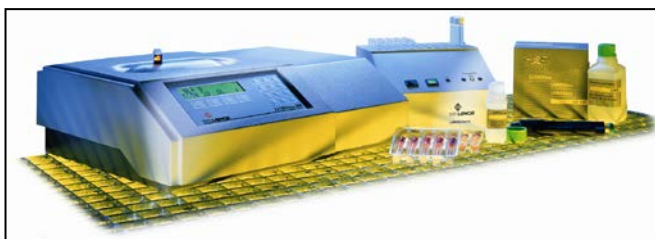
### Quality Assurance

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 LUMIStox luminometer is based on information provided by the vendor and does not represent verified information.

The LUMIStox 300 is a bench top luminometer that has been developed as a measuring unit for the luminescent bacteria test. In combination with the LUMIStherm incubation block it conforms to the technical requirements of ISO 11348-1, 2 and 3.



The LUMIStox 300 has a built-in photometer function and an automatic measuring and evaluation routine, which enables it to recognize color effects in the luminescent bacteria test and to take these into account in the test results.

The LUMIStox 300 can be connected to a PC running LUMISsoft4 that enables the operator performing and recording luminescent bacteria tests to conduct ISO 11348-1, 2 and 3 requirements. The results from LUMIStox are either LID or EC<sub>50</sub>-values, representing Lowest Ineffective Dilution (LID) causing less than 20%<sup>1</sup> inhibition and Effective Concentration (EC) causing 50% inhibition, respectively. The EC<sub>50</sub> values are a commonly used result from toxicity tests, while the LID is used in Germany. In this verification, results have been obtained for EC<sub>50</sub> values or directly as percentage of inhibition.

## VERIFICATION RESULTS

Table 3 Description of matrix and effect for LUMIStox.

Matrix	Effect
LUMIStox 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.  <b>Additional parameters included:</b> User manual quality, product cost, environmental health and safety.

### Performance Parameters

The performance parameters listed in Table 4 are results for EC<sub>50</sub> 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<sub>50</sub> results.

Table 4 LUMIStox performance parameter summary.

LUMIStox Compound	Criterion of detection	Range of application	Precision		Agreement with accepted values	Robustness		
			Repeat-ability	Reproduc-ibility		pH, color, turbidity, laboratory temperature <sup>1)</sup>	Cuvette material	Waste-water matrix <sup>1)</sup>
	% inhibition	mg/L	%	%	%	%	%	%
General	5.8							
Zn <sup>2+</sup>		8.3 -130	5.0	30	186		106 -117	43 -123
Cr <sub>2</sub> O <sub>7</sub> <sup>2-</sup>		35 - 560	29		91			0 -22
Triclosan		1.1 - 17	5.5		189			96 -141
Cyanide		48 - 780	24					
SDS		2.0 - 32	33			71 - 114	90 - 101	28 - 96
CTAB		1.9 - 31	2.4		100			68 - 78

<sup>1)</sup>For colored samples, robustness results are presented after the use of color correction. For the BaSO<sub>4</sub>-turbid samples, robustness results are presented without 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 -20% to 70%. The values listed in table are the best achievable robustness.

<sup>1</sup> LID of 20% inhibition is stated in ISO 11348-3, Annex B, Section B.5.

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 BaSO<sub>4</sub> 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 LUMIStox, LUMIStherm and LUMISoft4 software at the time of testing was 13,000 Euro (17,800 \$U.S.). Additional equipment such as cuvettes, bacteria and chemicals on a cost-per-sample basis as used for testing for EC<sub>50</sub> 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 EC<sub>50</sub> 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
<i>Capital</i>	
Buildings and land	1 laboratory facility
Equipment	1 LUMIStox, 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
<i>Operation and maintenance</i>	
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 energy	Power (PC and screen ~6 kWh, LUMIStox~0.4 kWh, LUMIStherm~0.4 kWh)
Labor	One laboratory technician for 1 day

Original signed by Mette Tjener Andersson 27/4-11  
 Verification responsible  
 Head of DHI DANETV Water Center

Original signed by Jørn Rasmussen  
 Director, Group R&D and Quality Management, DHI

Original signed by Tracy Stenner  
 Manager, Environmental Solutions Product Line  
 Battelle

Original signed by Sally Gutierrez  
 Director, National Risk Management Research Laboratory  
 Office of Research and Development, U.S. EPA

Original signed by Mona El Hallak  
 Director Technology Assessment  
 And Quality Assurance Services  
 OCETA (ETV Canada)

Original signed by Kevin Jones  
 President and CEO  
 OCETA (ETV Canada)

**NOTICE:** ETV verifications are based on an evaluation of technology performance under specific, predetermined operational conditions and parameters, and the appropriate quality assurance procedures. DANETV, DHI, ETV Canada, U.S. EPA and Battelle make no expressed or implied warranties as to the performance of the technology and do not certify that a technology will always operate as verified. The end user is solely responsible for complying with any and all applicable regulatory requirements. Mention of commercial product names does not imply endorsement.