The Baroxymeter: A Portable Respirometer for the Direct Toxicity Assessment of Wastewater

'To assess the possibility of exposure to contaminants that may not be detected by chemical monitoring, direct toxicity assessments should be considered as additional parameters for an adequate estimation of the water chemical status'. This abstract from the EU Water Framework Directive advocates the use of bioassays for the assessment of toxicity in waters and wastewaters for the simple reason that chemical analysis does not and cannot give direct information on their toxic content..

The regulation of industrial effluents has been based on the identification and control of specific substances and properties of the effluent such as pH, BOD, certain chemical contaminants etc. Although many discharges may be adequately controlled in this way, some complex effluents need additional control measures to ensure an appropriate level of environmental protection. Also, the detailed chemical analysis of complex effluents may often be prohibitively expensive. Direct toxicity assessment (DTA) is a risk-based approach using effects on biological systems as a basis for assessing likely harm to ecosystems in waters receiving effluent discharges. DTA is mainly aimed at complex effluents, where the combined effect of all substances present and the nature of their interactions are unknown. DTA is thus an index of the toxicity of the entire effluent, rather than a measurement of the concentration of specific effluent substances. It can provide an integrated assessment of the combined biological effects of all constituents, including unknown substances and those for which environmental quality standards or analytical methods do not currently exist. DTA can predict risk to the environment and assess the effectiveness of process control measures. As such, it is usually concerned with estimating the acute lethal toxicity of effluents using a suite of standardised aquatic ecotoxicity tests. In general, DTA is complementary to traditional chemical-specific analysis of effluents.

There are a large number of assays and instruments for analysis of wastewater and fresh water and, more specifically, for the detection of pollutants. However, most analytical instruments that can determine the toxicity of waters are laboratory-based, typically produce results only after days or hours, and in most cases require specially trained personnel. In cases where a toxicity assessment is rapidly needed, for example in a wastewater treatment plant influent (which, if toxic, may inhibit the biological activity of the activated sludge population of the plant with clear ecological and financial consequences) a method or an instrument that can rapidly detect a toxic effect is of paramount importance, not only for detection but also to assess possible protective actions. At the moment, regulators and industry appear to agree that DTA should include a suite of monitoring techniques, including a portable, rapid-response indicator to detect the presence of toxicity in influents and effluents. The latter, as a low-cost portable system that can be used in the field for incident detection, troubleshooting, and policing of potential problem discharges, could be an essential member of the monitoring suite.

Figure 1. The Baroxymeter: a portable respirometer for the detection of toxicity in wastewater.

The Baroxymeter *(Figure 1)*, developed by Biosynergy (Europe) Ltd. and marketed in the UK by TCS Biosciences Ltd, is a portable respirometer designed for the direct toxicity assessment of wastewaters. The Baroxymeter is aimed at the busy operator in need of an early rapid toxicity warning system that can be used directly with the sample of interest. The role of the Baroxymeter is to protect the activated sludge in a wastewater treatment plant from a toxic influent. It can be used by the wastewater treatment plant operative to test the influent for toxicity. In addition, the waste producers themselves (e.g. pharmaceuticals, chemical manufacturers, food plants) can test to ensure that their waste meets toxicity consents before release.

Principle

The instrument monitors the respiration of a Pseudomonas putida culture in the presence of a test effluent sample, relative to a reference sample, and reports the result as percentage respiration inhibition. This is based on the principle that a toxic sample will inhibit bacterial respiration. Respiration is measured by monitoring pressure drop over time in the headspace of a closed chamber containing the culture in the presence of the effluent sample. A soda lime cartridge absorbs carbon dioxide generated during respiration, so the measured pressure change is due principally to oxygen consumption by the culture.

System Elements

The Baroxymeter kit includes Ps. putida culture vials; toxic and non-toxic calibration samples; a soda lime cartridge; software for data processing and operational control. The instrument displays toxicity as a numeric value, and an additional green-amber-red light display gives an immediate 'toxicity' rating.

Operational overview

The operation of the Baroxymeter is conveniently simple. A sample is placed in the test chamber and the lid is closed. The test programme takes 10 minutes *(Figure 2)*. The sample is incubated for 5 minutes, and then undergoes a series of test cycles. Each cycle comprises a 30-second vent interval (chamber open to atmosphere) and a 30-second measurement interval (chamber sealed).

The instrument is calibrated on-site using the reagents provided. Thereafter, tests can be carried out at the site without further calibration. On completion of a test, the Baroxymeter indicates the toxicity result as a traffic-light LED display where green = non-toxic, amber = suspect, and red = toxic. If the sample is 'suspect', a retest may be carried out.

Baroxymeter settings

At the default setting, respiration inhibition of ≥40% is toxic, whereas ≤20% inhibition is non-toxic. Results between these values are borderline, and further testing should be carried out. These toxicity thresholds are recommended, however, there is a facility to adjust the settings.

Data processing

The Baroxymeter can store up to 100 test results. The unit can be preloaded with site/sample names and operators' details. Also, there is a setting for evaluating a series of sample dilutions (10-100%), enabling

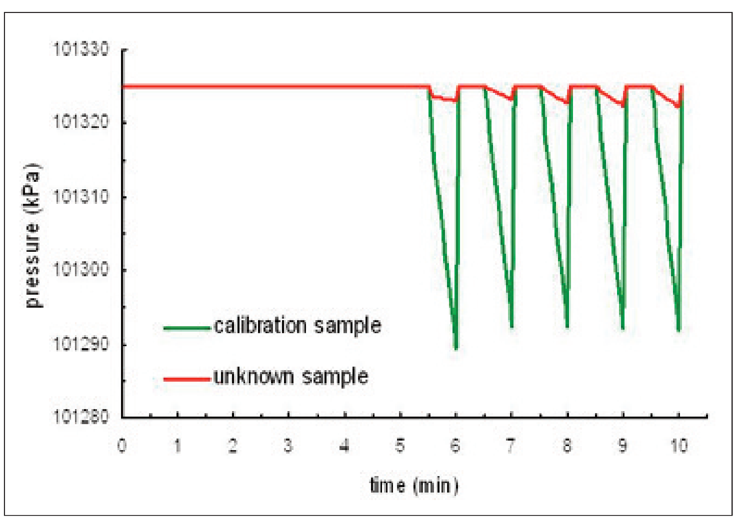

Figure 2: A typical Baroxymeter graph.

EC50 or other studies to be performed easily. There are three levels of software, enabling simple Excel downloads or complex data analysis, depending on the needs of the user.

Performance

Figure 3 shows results of an inter-laboratory reliability study conducted at the UK Environment Agency and Biosynergy (Europe) Ltd. An excellent agreement was demonstrated when testing the standard toxicant, zinc sulphate, using the Baroxymeter. These tests are part of a larger collaborative study.

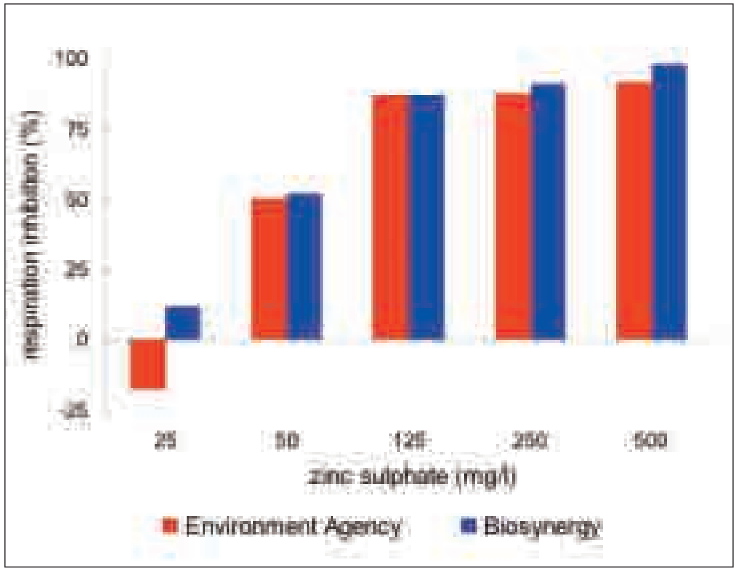

Figure 3: Respiration inhibition by zinc sulphate: inter-laboratory reliability study.

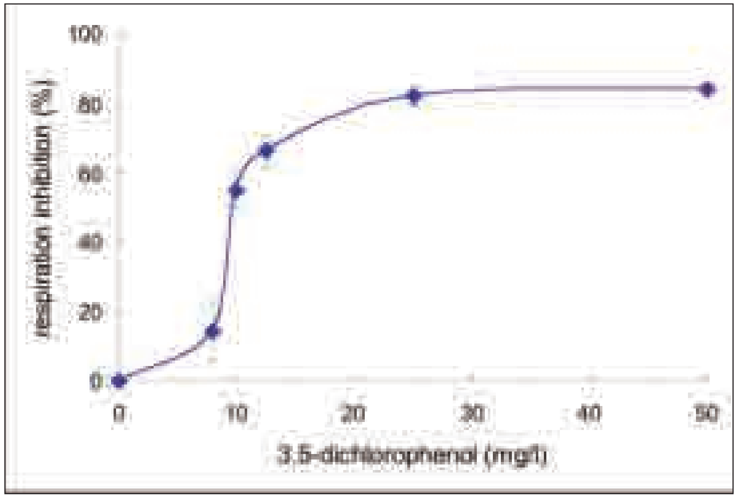

Figure 4. Dose-response curve for 3,5-dichlorophenol toxicity using the Baroxymeter.

Sample	Dilution	Respiration inhibition (%) (BAROXYMETER)	Result (BAROXYMETER)	Result (STRATHKELVIN)
B 22231	0	0	PASS	PASS
B 22232	0	15	PASS	PASS
B 22237	0	100	TOXIC	TOXIC
B 22237	1:10	0	PASS	PASS
B 22247	0	110	TOXIC	TOXIC
B 22247	1:10	22	PASS	PASS
B 22246	0	0	PASS	PASS
B 22039	0	71.5	TOXIC	TOXIC
B 22039	1:10	4	PASS	PASS
B 22202	0	14	PASS	PASS
B 22191	0	73.8	TOXIC	TOXIC
B 22191	1:10	0	PASS	PASS
B 22097	0	93.7	TOXIC	TOXIC
B 22097	1:10	0	PASS	PASS
B 22080	0	0	PASS	PASS

Table 1: Baroxymeter vs. Strathkelvin respirometer. Comparative toxicity testing results

A dose-response curve of 3,5-dichlorophenol (DCP) obtained with the Baroxymeter is shown in *Figure 4*. By testing serial dilutions of the initial 50mg/L sample, a dilution safe for disposal can be determined. In this test a concentration of DCP around 8mg/L was considered safe, and the EC50 is approximately 10mg/L.

Table 1 shows results from a comparative study between the Baroxymeter and the Strathkelvin respirometer, a laboratory-based instrument often used in toxicity studies. Detection of toxicity for both instruments is based on the monitoring of oxygen consumption by bacteria in contact with a sample however, the presentation of results is different for each instrument; hence in order to obtain comparable results, operators of both instruments reported results as pass or fail. The Baroxymeter was in complete agreement with the Strathkelvin respirometer not only in the detection of toxicity but also in determining safe dilutions.

Conclusions

Direct toxicity assessment is a risk-based approach using biological system tests to assess likely harm to ecosystems in waters receiving effluent discharges. The toxicity of the effluent as a whole is assessed, rather than its component substances. An instrument that can rapidly detect a toxic effect is of paramount importance, not only for detection but also to assess protective actions, in order to safeguard the wastewater treatment plant. The Baroxymeter is a portable respirometer designed for the direct toxicity assessment of wastewater. Toxicity can be detected within 10 minutes, on-site and by non-technical personnel. Studies have shown that the Baroxymeter can be used as a rapid early warning system for toxicity detection, and has the added advantages of being portable, inexpensive and easy to operate.

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