



# **HAB-BART system™**

*Technology Fact Sheet for Droycon Bioconcepts Inc.*

## **Performance Claim**

Droycon's HAB-BART system™ (heterotrophic aerobic bacteria – biological activity reaction test system) is a technology that allows the determination of heterotrophic aerobic bacteria (HAB) in water samples having a salinity of less than 4% salt. Quantification of respiratory HAB activity, when detected, is through the generation of a time lag to a reductive state in the tester and also qualitatively to differentiate the HAB into two major communities. The detection limit is equivalent to 67 cells per litre using a 15 ml water sample.

## **Technology Application**

The HAB-BART system™ has been used in the field and laboratory for the detection of HAB in waters suspected of having biofouling challenges. It forms a part of a strategy for the determination of the effectiveness of management practices in natural and engineered water systems. HAB are a significant bacterial component in many biofouling events that lead to compromised water systems with significant water quality and production problems.

## **Performance Conditions**

Any water sample taken for testing using the HAB-BART system™ must be collected following the protocols for the collection of a water sample for microbiological analysis. Transportation and storage of the sample should follow the standard guidelines practiced for sample handling prior to the initiation of microbiological examination. This should include aseptic handling, the use of sterile sampling containers and minimization of sample storage time to less than four hours at room temperature or twenty four hours when cooled to refrigeration temperatures. The HAB-BART system™ operates at  $28 \pm 1.0^\circ\text{C}$  and detects both mesophilic and facultatively psychrotrophic HAB. The packaged sterile tester has a four year shelf life when stored in a cool dry atmosphere. The HAB-BART system™ can be used for both laboratory and field based investigations and will generate similar data with respect to time lag and reaction patterns where a water sample is split and tested under similar conditions.

## **Technology Description**

The HAB-BART system™ uses three components which are the HAB-BART tester, HAB-BART reader and the BART-READ software (version H4). In this system the tester uses a test vial modified by the insertion of a floating ball and a dried pellet of selective chemicals documented to induce the growth of heterotrophic aerobic bacteria. To undertake a test using the tester, the water sample (15 ml) is added whereupon the ball floats up to create an aspect ratio conducive to the development of gradient that is oxidative near the ball and reductive below the ball. To initiate the testing the tester is inverted for thirty seconds to allow an indicator of the oxidation reduction (methylene blue dye) to dissolve into the sample. Five wrist actions while holding the tester now causes the sample to become saturated with oxygen from the headspace above the ball and the sample becomes blue in colour due to the oxidized state of the dye. The tester so prepared is now placed in one of the six accommodating slots in a HAB-BART reader. Two lateral 660 nm light pathways now pulse every second through the prepared sample in the incubating tester. While there remains oxygen in the sample, the dye remains blue but when there has been sufficient HAB respiratory activity in the sample then conditions turn reductive and the dye becomes colourless. This is detected by the affected light pathway through a sudden change in sorption from high (blue oxidized dye) to low (colourless reduced dye). The time lag (in seconds) between the start of the test and the detection of the sorption shift to reductive (colourless dye) can be used to quantify the population of respiring HAB in the sample on the basis that the length of the time lag (in seconds) is inversely related to the number of active cells within the sample being tested.



## Technology Description (continued)

BART-READ in conjunction with a suitable Windows compatible computer is now able to monitor the sorption shifts in the tester in real time and project a predicted active cells per ml (pac/mL) based upon a standard equation developed for HAB able to respire actively at  $28 \pm 1.0^{\circ}\text{C}$ . The level of HAB activity is recorded in two manners. First, the quantitative determination of the population achieved through the time lag (that period of delay in seconds) extrapolated by the standard formulation to pac/mL. Second, the manner in which the dye becomes reduced from the bottom of the tester upwards (UP) or from below the ball downwards (DO) differentiates two major HAB communities (aerobic or anaerobic respectively) either of which may dominate in any given sample. Under normal circumstances the test may last between 8 minutes to 5 days depending upon the size of the active HAB population.

## Verification

Droycon Bioconcepts Inc. carried out extensive examinations for HAB and the HAB-BART system™ test method that have been used independently by the Universities of Waterloo and Saskatchewan while the testers have been employed by Canada Agriculture and Agri-Food (Prairie Farm Rehabilitation Administration), the U.S. Army Corps of Engineers (sponsored by the Centers for Expertise, Omaha, Nebraska), and Leggette, Brashears & Graham Inc., Trumbull, Connecticut study of water well rehabilitation sponsored in part by the American Water Works Association Research Foundation, Denver, Colorado. The evaluation of the HAB is challenging, because there has not been a convenient field applicable test procedure that could be employed by operators who did not have extensive skills, knowledge and understanding of the microbiological examination methods for these bacteria. In recent times the HAB have become recognized as a major bacterial community of concern where they are detected in bodies of water. These concerns relate to the many aspects of biofouling including plugging, taste and odour quality problems and certain corrosive processes, but are also potentially significant in hygiene related issues and aerobic bioremediation processes. For these applications, the HAB-BART system™ has been shown to be a test that would perform effectively at the quantitative level differentiating two major HAB communities on the basis of the generated time lag and reaction patterns generated.

## What is the ETV Program?

The Environmental Technology Verification (ETV) Program is a joint Environment Canada - Industry Canada initiative delivered by ETV Canada Inc. The ETV Program is designed to support Canada's environment industry by providing credible and independent verification of technology performance claims.

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