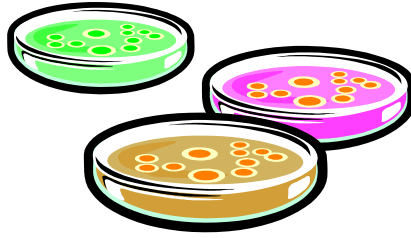


Comparison of BART™ Testers with Other Microbiological Tests

BART testers can use the water sample directly or they can accommodate solid or semi-solid samples. No dilution is employed and the tester is commonly incubated at room temperature although 28°C is faster and generates a greater precision. Where a liquid sample is used the tester accommodates 15mL for the test and the examination is for any activity or reactions within that tester caused by any of the microorganisms able to grow in the sample. For the solids or semi-solid samples (such as soils and muds), 0.1 to 0.5g are used diluted into a sterile diluant. In these tests the sample is not subjected to any dilution and so minimizes the impact on the microorganisms in the sample and improves the potential for effective recovery. Within the BART tester a variety of environments are established to increase the potential for activity and reactions to be observed.

Agar Spread Plates, Comparison with the BART testers



Spread plates use agar to make a jelly-like base upon which the bacteria can grow to form colonies (visible piles of cells) that are often easy to count and, with the right agar, can also be used to identify the types of bacteria that are present. That is why the counts using agar refer to “colony forming units” also known as “c.f.u”. The problem with the agar plates is that the microorganisms have to grow on the agar into a recognizable form commonly as a distinctive colony. While this method has a lot of convenient features it does not detect the many types of bacteria that are not able to grow on agar surfaces, cannot tolerate the high levels of oxygen, or cannot extract water effectively from the agar. BART testers have the advantage in that many more types of microorganism can grow in the BART tester often much faster because of the greater variety of environments that the tester presents to the organisms in the sample. BART testers are therefore more sensitive to a wider variety of microorganisms and can generate shorter delays before growth occurs. Therefore BART testers are **more sensitive** and **faster** than comparable agar spread plate techniques.

Agar Dip Paddles, comparisons with the BART testers

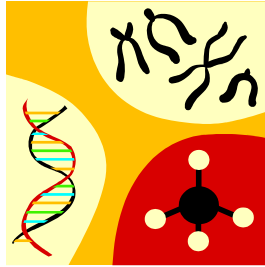
Dip paddles are constructed using a relatively thin agar film over a plastic paddle that is immersed in the water sample to inoculate the agar surface with microorganisms from the sample. Challenges for this technique are that the microorganisms have to become attached to the agar surface and then subsequently grow to form visible colonies when the dip paddle is incubated. Problems with this technique are that the agar forms a fairly thin film over the plastic and can begin to dry out quickly thus increasing rising the concentration of the chemicals in the agar and reducing the range of microorganisms that would effectively form colonies. BART testers, in using 15mL of sample, have a greater potential to cultivate such bacteria efficiently and therefore produce more precise data without having to be concerned about the agar drying out and giving inaccurate results.

Quick Tests (Color Change), Comparisons with the BART testers



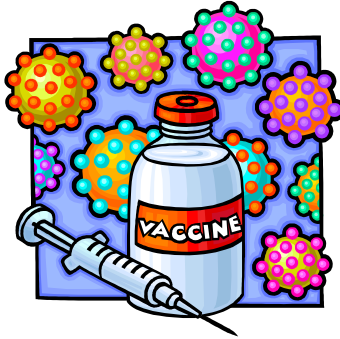
There are a range of fast microbiological tests that claim to detect the numbers of bacteria in a semi-quantitative manner (e.g., a lot, a few, and none). Such tests usually involve filtering the sample in order to trap the microbial cells that are then stained by some colored chemical reagent that reacts to these cells. The more cells present on the filter then the more intense the color. The problem with these techniques is that filtration means concentrating all of the organics (live and dead) onto the filter. The reagent may become reactive to the dead organic matter and signal a falsely high color reaction beyond the actual number of active cells present. While these tests are fast they lack the precision of the BART tester. Some BART testers such as the HAB-BART™ tester can react within a matter of 3,000 to 5,000 seconds when there are very large active populations of bacteria present (such as in primary influent to a sanitary waste water treatment plant) and can be reproducible. BART testers are quick tests too when there is a very large and active bacterial population in the sample.

ATP



All living cells produce adenosine triphosphate (ATP) as the main means to store energy in the cell. When a sample contains many active microbial cells then there is much more ATP present. Testing for ATP in a sample has become the “gold standard” by which the numbers of active cells in the sample can be counted. BART testers use a similar approach but here the activity is measured by the time lag to a recognized reaction or activity rather than the concentration of ATP. Here a shorter time lag would mean more activity and higher ATP levels. Studies at the Universities of Western Ontario and Saskatchewan has found good correlations between the concentration of ATP in water and soil samples and the time lags generated by BART testers. BART testers are, however, simpler to set up than the ATP analysis, less expensive and can also be used away from a laboratory setting. Time lags taken from the BART tester can provide good correlations to the ATP analysis but are more economical and more convenient in their use while at the same time obtaining good precision.

Immunoassay Tests, Comparison to the BART testers.



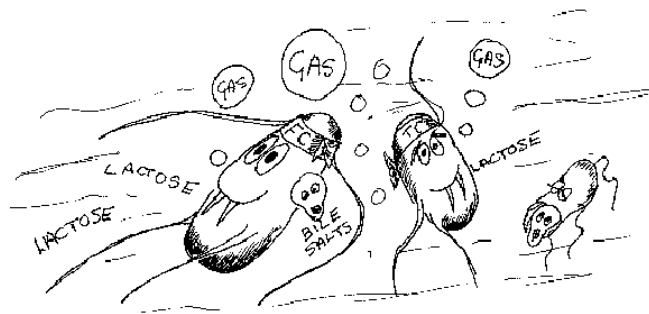
A whole new science called immunology was developed when it was realized that microorganisms all developed unique chemical signals. Some of those signals were specific to disease producing microorganisms and caused a specific response in the body enabling the body to become resistant (immune) to the pathogen. Today a whole biochemical science has evolved so that many microorganisms can be very precisely identified by unique chemical signals carried by the cells of a specific species. Some of these chemicals are called antigens and the infected body produces antibodies that neutralize them. By applying these antibodies wearing color coded tags it is possible to detect specific species of bacteria. These tests are more effective in a laboratory setting with highly trained technologists, however, some field tests do exist although there are often many precise steps in conducting the test. For example, an immunoassay test for sulfate reducing bacteria can involve six or more steps, precise timing and various apparatus to achieve a result. The highly precise technique used in the field test opens the door for a variety of errors to occur, resulting in false positives, or negatives. BART testers examine samples for whole communities of bacteria looking primarily for the quantification of the activity levels (population size) and reactions that are achieved (qualitative determination of the types of bacteria present). In simple terms the immunoassay tests are only better than the BART testers when the need is to detect a specific species of bacteria in the sample. In most samples there are active communities present that can include as many as eight to sixteen species just in each of those communities and for these conditions the BART testers offer more information with a simpler test method. Immunoassay tests are generally both complicated and expensive while the BART testers are simple, economical and can detect the nuisance bacteria causing the problems.

BOD, Comparison with the BOD-BART system

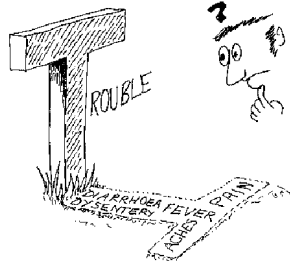


BOD stands for “biochemical oxygen demand” although some prefer to consider it as a “biological oxygen demand”. In discharging treated waste waters into the environment it is very important that these effluents do not trigger a sizeable oxygen demand in that environment. Such a demand could kill most of the animals including fish as the oxygen is consumed by the microbes in the effluent. Today the standard test remains a very widely recognized five day BOD test that has stood the test of time and is accepted to be functionally accurate. In the BOD-BART™ system oxygen demand is measured in the sample being tested by the time lag (in seconds) it takes for all of the oxygen to be removed to cause the blue color (oxidative) to disappear (reductive, oxygen is absent). This system can quantify the BOD in 3,000 to 60,000 seconds with precision and good correlations which have been obtained comparing the time lag to the five day BOD. This means that the test can be completed in less than 60,000 seconds compared to 432,000 seconds per test with the technologist time involved per test dropping the time spent actually working on the test to under five minutes for the BOD-BART system as compared to 30 to 90 minutes for the five day BOD depending upon the sample and the skill of the technologist. BOD-BART system is only recommended for the examination of sanitary municipal waste water treatment plants using either aerated lagoon or activated sludge treatment methods. BOD-BART system offers an 80% saving in testing time (at least) and an 80% saving in technician time as well as material costs. By using the BOD-BART system, the test is all over in less than 20 hours instead of 5 days with comparable precision.

Coliform Tests, Comparison with the T-COLI-BART system.



Coliform bacteria are defined by the fact that many of them inhabit the intestine; cause a range of diseases from diarrhoea, to dysentery, typhoid fever and a range of other infections. Since the start of the twentieth century, the presence of coliform bacteria in water has been considered to indicate that there is a hygiene / health risk to the user of that water. Testing for these bacteria has become more sensitive than for other bacterial groups and there is now commonly a zero tolerance for even one coliform bacterium in 100mL of water. There a number of tests that are in common use involving selective chemical that generate colors when the coliforms are active, the generation of colonies of distinctive types when grown on agars or filter materials, and the generation of gas. This latter technique was common through to the 1980s but has now been superseded by sensitive color reactions. All of these tests take 24 hours and commonly the use of blood heat for incubation. T-COLI-BART™ system uses the fermentation approach to detecting the coliform bacteria but without the dilution (a feature of the most probable number method, MPN). Here the 100mL sample is added directly to the tester which is then incubated in a reader. When gas is produced a dense plastic device floats up buoyed by the gas. As the device (D thimble™) floats up produced by the coliform bacteria in the sample. it triggers an infra red light that then determines the time lag in seconds. The larger the coliform population in the sample the sooner the D thimble comes up (normally in 20,000 to 80,000 seconds). Time taken to set up the test is less than five minutes and the operator is informed as soon as a positive detection has occurred. Other tests commonly have to be in a laboratory setting with careful controls. T-COLI-BART is a system that can be used in the field by trained certified operators and generates results in less than one day. Here the tester is actually recycled by return to DBI for sterilization and recycle.























Comparing the BART testers to other test methods

Many factors affect the use of a microbiological test and the comparison here covers the ease of use, cost of materials (remember that laboratories have many more costs than just materials to drive up the final costs), precision and operators time. These are summarized below for comparing the BART test methods in a general way to the other tests discussed.

Test	Ease of use	Cost (\$ materials per test)	Precision (± % variation)	Operators time (minutes)
Agar Spreadplate	Needs extensive training	\$1 to \$5	±80%	10
Agar Dip- Paddle	Little training required	\$2 to \$8	±80%	5
Rapid Test Methods	Little training required	\$1 to \$10	±100%	5 - 30
ATP Test Methods	Needs extensive training	\$15 to \$35	±10%*	10 - 120
Immunoassay Methods	Needs extensive training	\$15 to \$100	±10%**	15 to 90
5 day BOD test	Needs extensive training	\$5 to \$25	±20%	30 to 120
Coliform Test Methods	Needs extensive training	\$5 to \$100	Very variable depending on method	5 to 30
BART test Methods	Little training required	\$6 to \$12	±30%	5 to 15

Note: * indicates that the ATP can have very good accuracy when performed in the laboratory following standard procedures but some of the field ATP tests are crude to indicate whether living cells are present or not and these have significant potential variability; ** relates to the immunoassay techniques which can, in the laboratory setting using specific types of prepared samples, generate a high precision but might not be the case for tests that are used in the field setting.

Comparisons of the suitability of the tests for use by operators with a minimum of training is given below showing check marks

Test	Ease of use	Cost (\$ materials per test)	Precision (\pm % variation)	Operators time (minutes)
Agar Spreadplate	 extensive training	 \$1 to \$5	 $\pm 80\%$	10
Agar Dip-Paddle	 Little training	 \$2 to \$8	 $\pm 80\%$	 5
Rapid Test Methods	 Little training	 \$1 to \$10	 $\pm 100\%$	 5 - 30
ATP Test Methods	 extensive training	 \$15 to \$35	 $\pm 10\%*$	 10 - 120
Immunoassay Methods	 extensive training	 \$15 to \$100	 $\pm 10%**$	 15 to 90
5 day BOD test				



X by a characteristic means that the test method offers some serious drawbacks relating to price, precision, operator demands for training and time to perform the test.

From the chart it can be seen that the BART offers a convenience of ease of use, low price per test, precision and less operator time to set up the test which offers particular advantages when undertaking the tests away from a laboratory. It should be remembered that precision can be improved when using the BART systems which allow laboratory level accuracy when testing samples with good precision levels being achieved.

February 6, 2005 H:/general BART data 0804/comparison of BART with...