For the Detection of Problem-Causing Bacteria Involved in Microbiologically Influenced Corrosion (MIC)



# INSTRUCTIONS

Read all instructions before sample collection

*Before collecting sample*, review contents of this test kit (page 3). This kit allows testing of *one* sample. You will need one test kit for each location you are sampling.

*Determine the type* of sample to be analyzed (e.g., water or material from the inside surface of a pipe), and follow appropriate instructions below.

Process sample immediately after collection.

*Do not contaminate* sample by touching with non-sterile tools or hands.

*Warranty information* can be found on page 3 of these instructions.

*Dilution series in Section 1D* can be changed to suit your needs. Contact us if you require assistance.

Important: Properly dispose of all testing materials. Needles must be destroyed before disposal by cutting or bending back the needle. Syringes must be destroyed by breaking or shattering the barrel. Federal and local laws apply.

# **SECTION 1. MICROBIOLOGICAL TESTS**

### 1A. Collection: Solid Samples (Nodules, Corrosion Product, Soil, Etc.)

- 1. Remove silver tear-off cap and grey rubber stopper from BTI-ADS bottle. *Be careful* not to touch inside of stopper or mouth of bottle with hands or tools.
- Use sterile tongue depressor to add a portion of sample—about <sup>1</sup>/<sub>2</sub> teaspoon—to bottle.
- 3. Replace rubber stopper.
- 4. Shake vigorously to homogenize sample. You have just created what is known as a *slurry*.
- 5. Proceed to step 1D-1.

## 1B. Collection: Samples of Surface Scale or Biofilm

- 1. Remove silver tear-off cap and grey rubber stopper from BTI-ADS bottle. *Be careful* not to touch inside of stopper or mouth of bottle with hands or tools.
- 2. Dip sterile cotton-tipped swab into solution in BTI-ADS bottle to wet swab.
- 3. Swab an area of approximately one square inch of surface to be sampled.
- 4. Place cotton tip of swab into BTI-ADS bottle. Break off wooden portion of swab touched by your fingers, and discard.
- 5. Replace rubber stopper.
- 6. Shake vigorously to homogenize sample. You have just created what is known as a *slurry*.
- 7. Proceed to step 1D-1.

## 1C. Collection: Liquid Samples

- 1. Remove silver tear-off cap and grey rubber stopper from BTI-ADS bottle. *Be careful* not to touch inside of stopper or mouth of bottle with hands or tools.
- 2. Discard solution into an appropriate waste container.
- 3. Fill BTI-ADS bottle with liquid sample.
- 4. Replace rubber stopper.
- 5. Proceed to step 1D-1.

### 1D. Inoculation of Media

- 1. Using a marking pen, label each bottle in each string (color) of media 1 through 5. Start with yellow-capped bottles.
- 2. Wipe rubber stopper of BTI-ADS bottle with alcohol prep pad. This will re-sterilize the stopper.
- Remove and discard wrappers from a sterile 1 ml syringe and an 18g needle. Without touching tip of syringe or opening of needle, place needle onto syringe. Tighten needle onto syringe by pushing in and turning needle shield clockwise.
- 4. Remove needle shield. Insert syringe needle through rubber stopper and into sample/slurry in BTI-ADS bottle.
- 5. Withdraw 1.0 ml of sample/slurry from BTI-ADS bottle by gently pulling up on syringe plunger until sample/ slurry reaches the 1.0 ml mark.
- 6. Flip plastic cap off first yellow-capped bottle (labeled #1).
- 7. Insert syringe needle through rubber stopper of first bottle. Inject sample/slurry into bottle by depressing plunger.
- 8. Keep needle in bottle. Mix solution in bottle by gently withdrawing plunger, drawing up **1.0 ml** of media-sample/slurry mixture, and then depressing plunger, reinjecting liquid into bottle. Repeat several times.
- 9. Withdraw **1.0 ml** of solution from yellow bottle #1 and inject into yellow bottle #2. Mix as in step 8.
- 10. Now, withdraw **0.1 ml (one-tenth!)** of solution from yellow bottle #2 and inject into yellow bottle #3. Mix as in step 8.
- 11. Withdraw **0.1 ml** of solution from yellow bottle #3 and inject into yellow bottle #4. Mix as in step 8.
- 12. Withdraw **0.1 ml** of solution from yellow bottle #4 and inject into yellow bottle #5.
- 13. Repeat steps 2 through 12 for the blue, red, and greencapped bottles using new 1 ml syringes and needles.

## SECTION 1. MICROBIOLOGICAL TESTS (CONT'D)

14. Keep all bottles of media in closed kit box at room temperature. Proceed to Section 2.

# **SECTION 2. INTERPRETATIONS**

### 2A. Interpretations of Results

After 2, 5, and 15 days incubation, compare microbiological test bottles to written descriptions, below, and to Positive Reactions Sheet, attached. Record results in attached Test Data Sheet, and record any changes.

- 1. Yellow-capped bottles detect the presence of aerobic bacteria. These bottles will turn cloudy and/or have slime, which appears as sheets or clumps of material, if aerobes (AERO) are present. Record highest bottle number to turn positive (1 through 5).
- 2. Blue-capped bottles detect anaerobic or facultatively anaerobic bacteria. These bottles will turn cloudy if anaerobes (ANA) are present. Record highest bottle number to turn positive (1 through 5).
- 3. Red-capped bottles detect organic acid-producing bacteria (APB). These bottles will turn cloudy orange or cloudy yellow if APB are present. Record highest bottle number to turn positive (1 through 5).
- 4. Green-capped bottles detect sulfate-reducing bacteria (SRB). These bottles will turn black or will have black slime form on the iron nail along the bottom of the bottle if SRB are present. The presence of black or gray flecks is **not** a positive reaction for SRB. Record the highest bottle number to turn positive (1 through 5).

# 2B. Calculating the Number of Bacteria in Your Sample

*NOTE:* You have performed serial, decimal dilutions of your sample in these media bottles. This means you can get an approximation of the numbers of viable bacteria in the sample using the chart below:

| Highest Bottle # to Turn Positive | Range of Viable Bacteria per mL of Liquid Sample |
|-----------------------------------|--|
| 1                                 | 1 to 10  |
| 2                                 | 10 to 100  |
| 3                                 | 1,000 to 10,000                                  |
| 4                                 | 100,000 to 1 million                             |
| 5                                 | ≥10 million                                      |

| Highest Bottle # to Turn Positive | Range of Viable Bacteria<br>per gram of Slurry |
|-----------------------------------|--|
| 1                                 | 10 to 100                                      |
| 2                                 | 100 to 1,000                                   |
| 3                                 | 10,000 to 100,000                              |
| 4                                 | 1,000,000 to 10 million                        |
| 5                                 | ≥100 million                                   |

### 2C. Formal Interpretation

For a formal written report on results, interpretation, conclusions, and recommendations, return completed Analytical Requests Sheet (attached) and completed test kit to BTI Products. An additional fee is assessed for this service. Call 970.884.4629 for details.

For technical assistance, to request MSDS, or to place an order:

Call Toll Free: 970.884.4629

Or E-mail: products@bti-labs.com

# MICkit<sup>®</sup> 3: List of Kit Contents

- 5 Bottles BTI-AERO Medium (Yellow Flip-Off Caps)
- 2. 5 Bottles BTI-ANA Medium (Blue Flip-Off Caps)
- 3. 5 Bottles BTI-APB Medium (Red Flip-Off Caps)
- 4. 5 Bottles BTI-SRB Medium (Green Flip-Off Caps)
- 5. 1 Bottle BTI-ADS (Silver Tear-Off Cap)
- 6. 4, 1 ml Syringes
- 7. 4, 18g Needles
- 8. 1 Sterile Tongue Depressor
- 9. 1 Sterile Cotton-Tipped Swab
- 10. 1 Alcohol Prep Pad

# WARRANTY

**BTI Products, LLC's** products are warranted by **BTI Products, LLC** to perform as described in the technical literature supplied with each product, provided the products are used, stored, and maintained in accordance with the directions provided. They must also be used before the expiration date. Adequate quality control must be done by the user of the products.

**BTI Products, LLC** disclaims any implied warranty of merchantability or fitness of its products for any other purpose than described in its technical literature, and in no event shall **BTI Products, LLC** be held liable for any consequential damages arising out of the aforesaid express warranty.

Should you have questions about this product or any of the products and services we provide, please call or write:

BTI Products, LLC 652 Silver Hills Road Bayfield, CO 81122 970.884.4629 products@bti-labs.com

We welcome all comments and inquiries.

**Usage & Storage:** Use by expiration date printed on kit box label. Store test materials in a cool, dry place out of direct sunlight. Do not eat or drink any of the contents of the kit. Keep out of the reach of children. Material Safety Data Sheets available upon request.

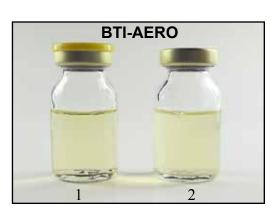
**Disposal of Test Materials:** Properly dispose of all kit components. Needles must be destroyed before disposal by cutting or bending back the needle. Syringes must be destroyed by breaking or shattering the barrel. Federal and local laws apply.

Used media bottles must be properly disposed of according to local regulations. Alternatively, bottles/kits may be returned to **BTI Products, LLC** for proper disposal for a fee of \$30.00 per kit.

**Need Help?** 

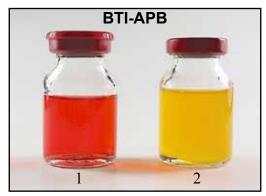
Call 970.884.4629

Rev 2/15/22



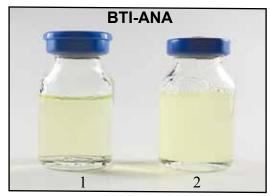
1. Uninoculated (Negative)

2. Positive—Cloudy with possible slime or fungus formation



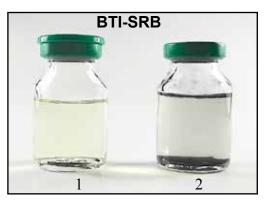
Uninoculated (Negative)
 Positive—Cloudy orange or yellow

-Slime formation without a color change does not indicate a positive



- 1. Uninoculated (Negative)
- 2. Positive—Cloudy

-An amber color change does not indicate a positive



1. Uninoculated (Negative)

2. Positive—Black color change -Positives can include black slime formation on iron nail



# MICkit<sup>®</sup> 3 - Positive Reactions Sheet

### ANALYTICAL REQUESTS SHEET Please fill out completely and return with the sample(s). Send to: BTI Products, LLC 652 Silver Hills Road Bayfield, CO 81122 970.884.4629 products@bti-labs.com

#### I. Sample Information

| <ol> <li>Sample name or site designation</li> </ol> |  |
|---|--|
| 2. Date sample collected                            |  |
| 3. Date sample shipped                              |  |
| 4. Type and location of sample                      |  |
| 5. Company name and address                         |  |
| 6. Contact name                                     |  |
| 7. Telephone and email                              |  |
| 8. PO or Credit Card #                              |  |
| 9. Name on card                                     |  |
| 10.Billing address                                  |  |
|   |  |

Please indicate below which analyses you wish to have performed on the sample. If you have any questions, please contact us at 970.884.4629.

### II. Sample Analyses

| А. | Microbiological Analyses |
|----|--------------------------|
|----|--------------------------|

| 1. Viable culture   | Cost Per Sample | Yes |
|---|-----------------|-----|
| <ul> <li>a. MICkit<sup>®</sup> 3 – Inoculated by client and read by BTI Products</li> <li>b. MICkit<sup>®</sup> 3 Inoculated and read by BTI Products</li> <li>2. Other (specify)</li></ul> | \$236<br>\$382  |     |
| B. Other  |                 |     |
| <ol> <li>Pipe analysis</li> <li>Photodocumentation</li> <li>Other (specify)</li> </ol>  | Quote<br>Quote  |     |

# **MICkit® 3 TEST DATA SHEET**

| Sample Information                          |  |  |
|---|--|--|
| Facility                                    |  |  |
| Sample Name or Designation                  |  |  |
| Test Date                                   |  |  |
| Type of Sample Tested                       |  |  |
| Microbiological Results                     |  |  |
| Aerobic Bacteria (per ml/per gram)          |  |  |
| Anaerobic Bacteria (per ml/per gram)        |  |  |
| Acid-producing Bacteria (per ml/per gram)   |  |  |
| Sulfate-reducing Bacteria (per ml/per gram) |  |  |