



## Serial Dilution Instructions (AnPRD) FOR DETECTION OF ANAEROBIC ACID PRODUCING BACTERIA AND GENERAL ANAEROBIC BACTERIA

- Product Description:

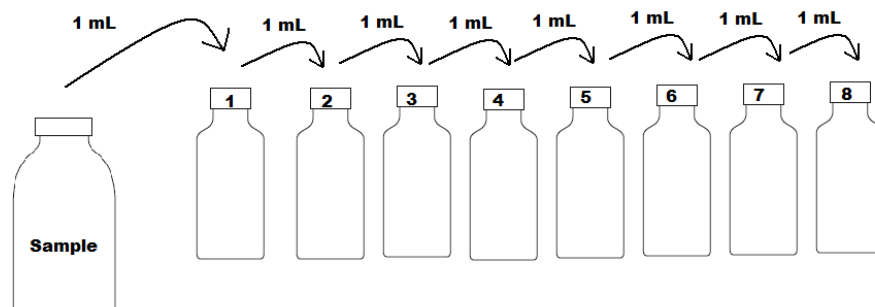
1. N.A.C.E. standard (TM0194-2004) Anaerobic Phenol Red Dextrose Media
2. Anaerobic bacterial growth media, such as Anaerobic Phenol Red Dextrose (AnPRD), is extremely sensitive to oxygen exposure. Biotechnology Solutions' anaerobic bacterial growth media is manufactured inside custom built anaerobic chambers to ensure the highest quality bacterial growth media products.
3. **Physical Characteristics:** Bright to Dark Red in Color
4. **Chemistry:** Contains proteins, sugars, and a pH indicator.
5. **Detection:** Used for the enumeration of total Anaerobic Acid Producing Bacteria(APB) as well as total General Anaerobic Bacteria(GAB)

- Collecting a Sample:

1. Collect the sample in such a manner as to preclude contamination from external sources.
2. Time, date, temperature, and appearance of the sample should be recorded.

- Preparation:

1. Arrange selected media vials into "Dilution Series".
2. The selected media should approximate the conditions (Temp., TDS, etc.) of the sample water being tested to avoid the "shock" effect on the microbes.



For additional information, please contact us at:  
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- Sterilization:

1. Wipe the rubber caps of the media vials with sterile alcohol pads.

- Inoculation:

1. Using a sterile disposable syringe, withdraw 1 mL of the sample and inject it into bottle #1 and discard syringe. Mix contents thoroughly by vigorously agitating the vial. Some bubbles may appear; this is normal.
2. Make sure to note changes in the vial upon inoculation that may cause a **False Positive**.
  - If the vials turn yellow immediately upon injection, the color change is due to the low pH of the sample fluids, not biogenic activity. If the questionable positive media is perfectly clear even though the yellow color change has taken place, this is likely due to contamination and not bacterial activity. If acid producing bacteria are present in the vial, turbidity (cloudiness) of the media will be apparent visually. If you are not sure if the first vial turned immediately upon inoculation or what the pH of the sample is, you can subculture the vial in question by performing a serial dilution using 1 mL from that vial. We recommend having a water analysis done so that you know what constituents and at what concentration make up the sample.
3. With a new sterile syringe, withdraw 1 mL of solution from bottle #1 and inject it into bottle #2 and discard syringe. Mix contents thoroughly by vigorously agitating the vial.
4. Repeat this process for all the remaining dilution vials (#3 - #6).
5. Incubate the vials at the temperature at which the original sample was collected ( $\pm 2^{\circ}\text{C}$ ). N.A.C.E. standards call for 28-day incubation for SRB media and 14 days for APB media.

- Reading:

1. **Indicator for (APB):** Acid Producing Bacteria will drop the pH of the media causing a color change from red to yellow. The color change must also be accompanied by biomass or turbidity
2. **Indicator for (GAB):** General Anaerobic Bacteria can grow in this media as well but will not cause a color change from red to yellow. GAB can be identified in the vials by visually identifying turbidity or biomass accompanied by NO change in the red color of the media.
3. Record the number of positive vials.

- Disposal:

### How should we dispose of Biotechnology Solutions' microbiological media waste?

All of BTS bacterial growth media products are considered to be non-hazardous materials. Media vials may be discarded according to your local, state and federal regulations. To find out more about these regulations please refer to the environmental, health and safety staff at your facility.

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