MicroFast[®] Coliform & E. coli Count Plate

MF1007

AOAC Validated (Certificate No.102103)

Introduction

MicroFast Coliform & E. coli Count Plate (EC) is a sample-ready-culture medium system. It uses innovative technologies such as rapid diffusion systems and new-generation microbial coloration to achieve rapid proliferation and interpretation of colonies, greatly improving the detection efficiency in the laboratory.

The plate contains prefabricated type of medium, cold-water gel and indicator (Triphenyl tetrazolium chloride). It is intended for the enumeration of coliform and Escherichia coli in food and production environments.

Certified to International Organization for Standardization (ISO) 9001 for design and manufacturing.

Sample Preparation

- 1. Use appropriate sterile diluents: i.e., sterile water or phosphate buffered diluent.
 - a) For liquid samples, the stock solutions can be used directly for inoculation.
 - b) For stainless steel surfaces, pre-moistened sampling sponge with 10 mL PB buffer.
 Sample each 100cm² test area by using firm and even pressure 10 times diagonally, vertically, and horizontally. After sampling, add 90mL volume of buffer and homogenize. Make 10-fold serial dilutions of the sample homogenate.

Note: do not use diluents containing citrate, bisulphite or thiosulphate with MicroFast plates as they could inhibit growth.

2. Blend or homogenize sample completely with 1 part sample and 9 parts diluent (sterile saline solution or sterile phosphate buffer) (1:10 dilution scheme).

For optimal growth and recovery of microorganisms, the pH pf the sample suspension should be adjusted to pH 6.5-7.5.

For acidic products, adjust the pH with 1N NaOH. For alkaline products, adjust the pH with 1N HCl.

3. Prepare decimal dilutions of the homogenized sample solution by transferring 1mL of the homogenized solution to a tube containing 9mL of sterile diluent and mix completely. Based on sample type, select 2 or 3 suitable diluted sample solutions that will result in the countable range for the MicroFast plate type being achieved.

Operation Procedure

- 1. Open the aluminum foil bag and place the MicroFast Plate on a flat, level surface.
- 2. Lift the top film whilst supporting the plate without touching the test area.
- 3. With the pipette vertical to the inoculation surface, dispense 1mL of sample suspension onto the center of bottom film.
- Drop the top film slowly onto the sample and the solution will spread automatically. Leave for at least one minute to allow the solution to spread completely before moving the plate to the incubator.



Incubation

Incubate MicroFast plates in a horizontal position with the film upwards in stacks of no more than 20. Culture at $36\pm1^{\circ}$ C for $24\pm2h$.

Interpretation

- 1. MicroFast plates can be counted visually using a standard colony counter or other illuminated magnifier. Escherichia coli count blue colonies with bubbles, Coliforms count blue and red colonies with bubbles.
- The approximate size of the circular growth area is 20cm². Estimates can be made on MicroFast Plates containing between 15-150 colonies by counting the number of colonies in the circular growth area.
- Alternatively, estimates can be made on MicroFast plates containing greater than 150 colonies by counting the number of colonies in two or more representative squares and determine the average number per square. Multiply the average number by 20 to determine the estimated count per plate.
- 4. High numbers of colonies on the MicroFast plates will cause the entire growth area to appear discolored. Record this result as too numerous to count (TNTC). Where a count is required, evaluate the performance of the next dilution. If there are obvious colonies in the next dilution and within the optimal counting range, they should be counted and recorded.

If there is no colony count, the count plate might have either been contaminated or sample matrix has negatively influenced microbial growth on the count plate.

5. Too many other colonies may cause the E. coli cannot be counted and recorded.



Normal counting range

E. coli/Coliform Gas generated colonies interpretation instructions

Colonies that produce bubbles (gas) may be interpreted as positive in the following examples $\hat{\boldsymbol{\varphi}}$

- 1) Two colonies may be connected to one bubble;
- (2) A colony can be connected with one bubble or 2-3 bubbles or multiple bubbles;
- ③ Bubbles can be generated around the colonies and are not connected to the colonies;

(4) Bubbles and colonies are in the same position, which may break the colonies and cause colonies to grow on the edge of the bubbles.

Bubbles caused by other reasons (not produces from colonies)

- (1) Bubbles will be generated due to improper operation. At this time, the bubbles are not connected
- with the colonies, and the bubbles are large and the edges are irregular;
- (2) Foam-rich samples will cause bubbles in the count plate.

Storage

- 1. The shelf life of count plate is 18 months. Use up within the shelf life. Lot number can be found on the pouch.
- The count plates components are sterilized. Unopened count plates should be stored at 2°C-8°C. Equilibrate the count plate to room temperature before use.
- 3. After unsealing, stick the pouch with adhesive tape or seal it with a sealing clip, store it in the dark at room or ambient temperature 15°C-25°C, and use it up within one month.
- When transporting or short-term storage, store the count plate at room temperature (18-30°C).

Scope of AOAC Validation

- Fresh raw ground beef (75% LEAN), sliced deli ham, raw whole cow's milk, fresh bagged mixed greens, frozen fish and stainless steel (18 GA gauge), 300 series, brush finish, NSF certified) were certified in the AOAC Research Institute *Performance Tested Methods*SM study.
- MicroFast EC demonstrated equivalent performance to the U.S. Department of Agriculture Food Safety and Inspection Service Microbiology Laboratory Guidebook 3.02 Quantitative Analysis of bacteria in Foods as Sanitary Indicators and to the U.S. Food and Drug Administration Bacteriological Analytical Manual (FDA/BAM) Chapter 4: Coliforms Plate Count reference methods.

WARNINGS & PRECAUTIONS

- The user should read, understand, and follow all safety information in the instructions before use.
- The MicroFast Count Plate should be disposed following procedures for infectious or potentially infectious products. User should wear appropriate personal protective equipment, including, but not limited to, protective disposable gloves, laboratory coats, and eye protection when handling samples and kit reagents, Wash hands thoroughly after handling specimens and reagents. It is the responsibility of each laboratory to handle waste and effluents produced according to their type and degree of hazardousness and to treat and dispose of them (or have them treated and disposed) in accordance with local, state, and federal regulations. Strict compliance with BSL-2 practices should be followed.
- Follow all product storage guidelines included in the insert. Do not use after the expiration date.
- MicroFast Count Plate testing should be done in a professionally equipped laboratory under the supervision of a skilled microbiologist. The user must train its staff on the current testing methods.
- MicroFast Count Plates have not been reported for application in industries other than food and environmental samples. Use within the suggested scope.
- Counting results of MicroFast Count Plates may not be the same as agar.
- MicroFast Count Plate have not been evaluated with all possible food products, food processes, testing protocols or with all possible microorganism strains.
- As a general precaution, clean the workstations with the disinfectant of choice (e.g., sodium hypochlorite solution, phenol solution, quaternary ammonium solution) before and after, in addition to having work areas separated for the following: media preparation, sample preparation, and indicator organism enumeration. Gloves and other personal protective equipment should always be used.
- Count plate may contain microorganisms that may be a potential biohazard. Follow current industry standards for disposal.
- Keep the count plate away from ultraviolet, direct sunlight and fluorescent lamp.
- Do not use the polluted or damped count plate.

- If the pH of the test sample is too high or too low, it will affect the accuracy of the test results.
- When uncovering the film, do not touch the culture area of the medium.
- If there are too many colonies, the detection of positive strains might be affected.
- The count plate may show a few needle-like black spots, which is normal and does not affect the interpretation of the target strain.
- If the sample is viscous, diffusion can be aided manually.
- When pipetting samples, do not touch culture area.

Limitation of Warranties

Accurate results depend on the proper use of the kit by following the instructions for use carefully. If the kit fails to perform according to specification, please contact your sales representative at Scigiene.

Limitation of Scigiene Liability

Scigiene will not be liable for any loss or damages, whether direct, indirect, special, incidental or consequential damages, including but not limited to lost profits. In no event shall Scigiene's liability under any legal theory exceed the purchase price of the product alleged to be defective.

User Responsibility

Users are responsible for becoming acquainted with product instructions and information. For further information, please contact your local Scigiene dealer or distributor.

When choosing a test method, please note that external factors such as sampling methods, testing protocols, sample preparation, handling, and laboratory technique can all have an impact on the results.

When selecting a test method or product, it is the user's duty to assess a sufficient number of samples with the proper matrices and microbiological challenges to ensure that the chosen test methos meets the user's criteria. It is also the user's obligation to ensure that any test methods and results fulfill the criteria of its customers/suppliers.

Results acquired from the use of any Scigiene product, like any other method, cannot guarantee the quality of the tested matrices or processes.



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