

3M™ Petrifilm™

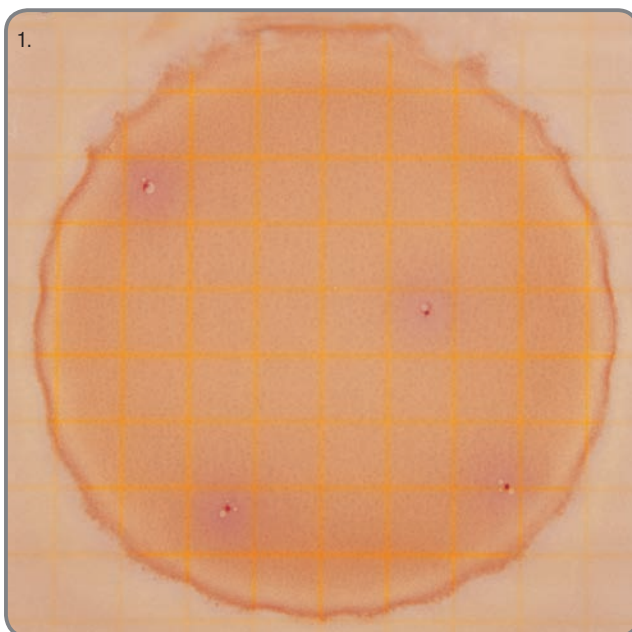
Interpretation Guide

3M™ Petrifilm™ High-Sensitivity Coliform Count Plates

This guide will familiarize you with results on 3M™ Petrifilm™ High-Sensitivity Coliform Count (HSCC) plates. For more information, please contact the official 3M Food Safety Products representative nearest you.

Petrifilm HSCC plates contain a ready-to-use modified Violet Red Bile (VRB) selective culture medium, a cold-water-soluble gelling agent and a tetrazolium indicator that facilitates colony enumeration. The top film traps gas produced by the lactose fermenting coliforms. Time and temperature of incubation as well as interpretation vary by method.

- **ISO** defines coliforms by their ability to grow in method-specific, selective media. **ISO method 4832**, enumerating coliforms by the colony count technique, defines coliforms by colony size and acid production on VRB with lactose (VRBL) agar. On Petrifilm HSCC plates, these acid-producing coliforms are indicated by red colonies with or without gas (within approximately one colony diameter). **ISO method 4831**, enumerating coliforms by the Most Probable Number (MPN) method, defines coliforms by their ability to grow and produce gas from lactose in a selective broth. On Petrifilm HSCC plates these coliforms are indicated by red colonies associated with gas (within approximately one colony diameter).
- **AOAC INTERNATIONAL** and U.S. Food and Drug Administration Bacteriological Analytical Manual (BAM) define coliforms as gram-negative rods which produce acid and gas from lactose during metabolic fermentation. Coliform colonies growing on the Petrifilm HSCC plate produce acid which deepens the gel colour; gas trapped around coliform colonies (within approximately one colony diameter) indicates coliforms.



The Petrifilm HSCC plate is designed for the detection of both total coliform and thermotolerant (faecal) coliform.

These Petrifilm HSCC plates are highly recommended to detect low numbers of coliforms in all foods.

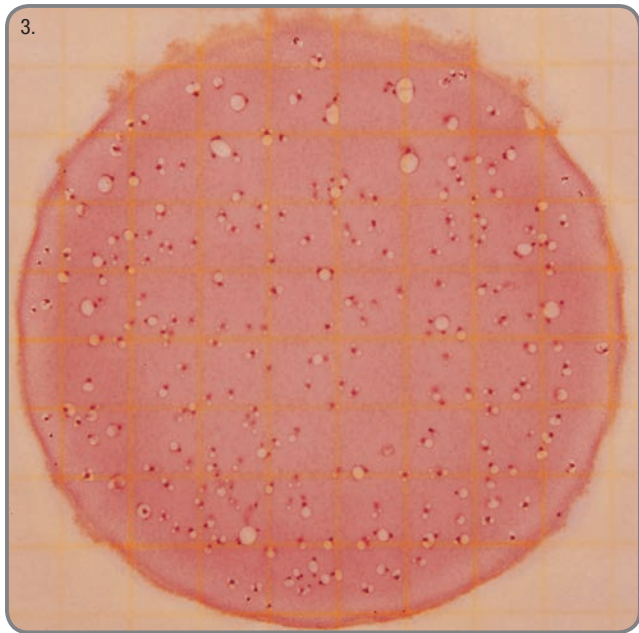
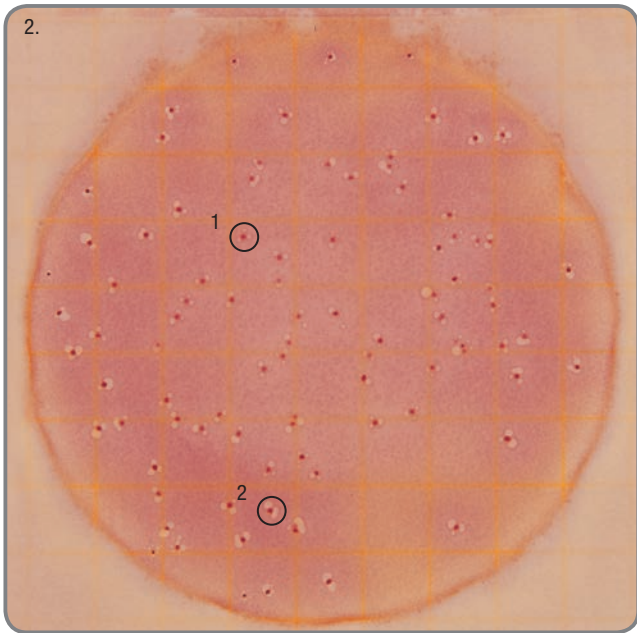
AFNOR have validated the use of Petrifilm HSCC plates under specific conditions. See Reminders for Use section of this Interpretation Guide.

Gas producing colonies count: 4



3M™ Petrifilm™ High-Sensitivity Coliform Count Plate

Notice the change in gel color in figures 1 through 5. As the coliform count and the production of acid increase, the color of the gel turns from light-orange to bright pink-red.



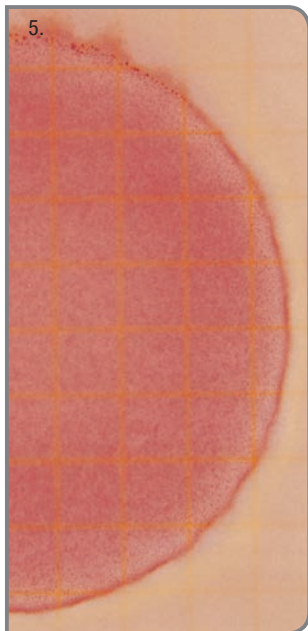
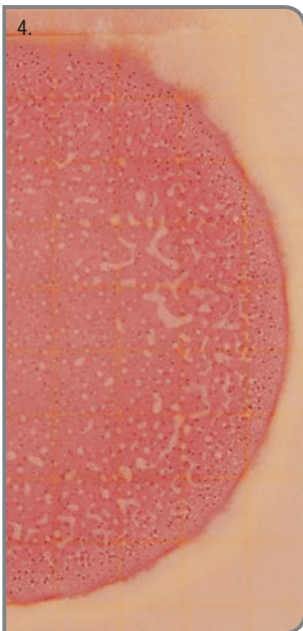
Gas producing colonies count: 82
Non-gas producing colonies count: 8
Total count: 90

Bubbles pattern can vary: see circles 1 and 2.
 Sometimes, the gas produced disrupts the coliform colony so that the colony "outlines" the bubble.

Estimated total count: 320

The circular growth area is approximately 60 cm². Estimates can be made on plates containing greater than 150 colonies by counting the number of colonies in one or more representative squares and determining the average number per square. Multiply the average number by 60 to determine the estimated count per plate.

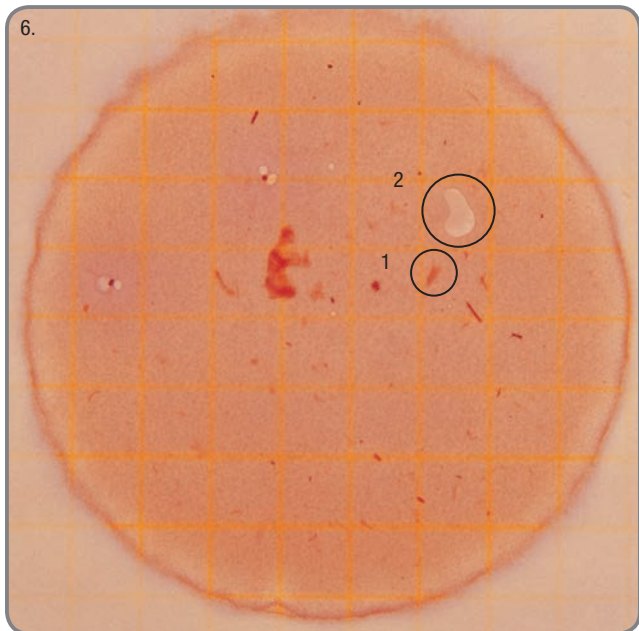
Further dilution of the sample is recommended to determine an accurate count.



TNTC (Too Numerous To Count) plates

Petrifilm HSCC plates with colonies that are TNTC have one or more of the following characteristics: many small colonies, many gas bubbles, deepening of the gel colour.

Further dilution of the sample is recommended to determine an accurate count.



Gas producing colonies count: 2

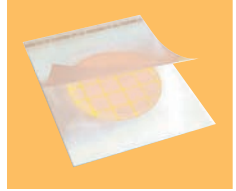
Food particles are often irregularly shaped and are not associated with gas bubbles (see circle 1).

Artifact bubbles may result from improper inoculation of the Petrifilm HSCC plates. They are irregularly shaped and not associated with a red colony (see circle 2).

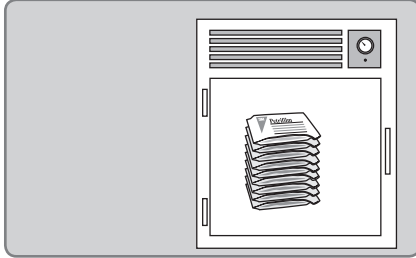
3M™ Petrifilm™ High-Sensitivity Coliform Count Plate

For detailed WARNINGS, CAUTIONS, DISCLAIMER OF WARRANTIES / LIMITED REMEDY, LIMITATION OF 3M LIABILITY, STORAGE AND DISPOSAL information, and INSTRUCTIONS FOR USE see product's package insert.

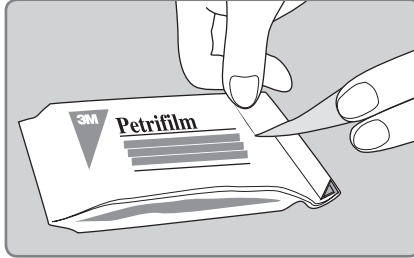
Reminders
for use



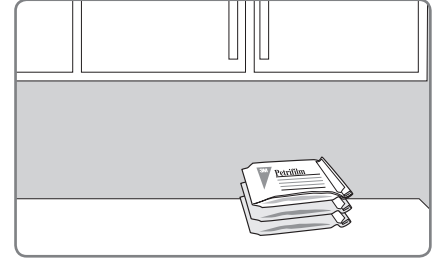
Storage



1 Store unopened packages at $\leq 8^{\circ}\text{C}$ ($\leq 46^{\circ}\text{F}$). Use before expiration date on package. In areas of high humidity where condensation may be an issue, it is best to allow packages to reach room temperature before opening.



2 To seal opened package, fold end over and tape shut.

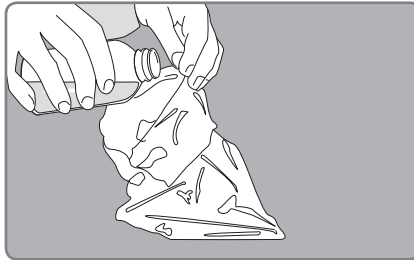


3 Keep resealed package at $\leq 25^{\circ}\text{C}$ ($\leq 77^{\circ}\text{F}$) and $\leq 50\%$ RH. **Do not refrigerate opened packages.** Use Petrifilm plates within one month after opening.

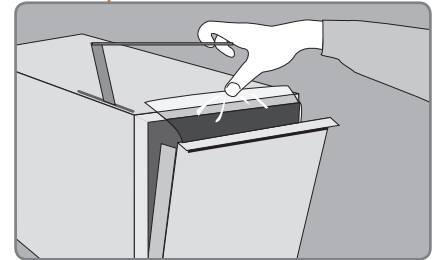
Sample Preparation



4 Weigh or pipette food product into an appropriate sterile container such as stomacher bag, dilution bottle, Whirl-Pak® bag, or other sterile container.



5 If necessary, use appropriate sterile diluents: peptone salt diluent or maximum recovery diluent (ISO method 6887), Butterfield's phosphate buffer (IDF phosphate buffer, KH_2PO_4 at 0.0425 g/L, adjusted to pH 7.2), 0.1% peptone water, buffered peptone water (ISO method 6579), saline solution (0.85 - 0.90% weight to volume), bisulphite-free letheen broth or distilled water.



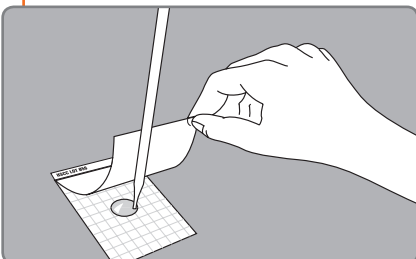
6 Blend or homogenize sample as per current procedure.

Adjust pH of the diluted sample between 6.5 and 7.5 :

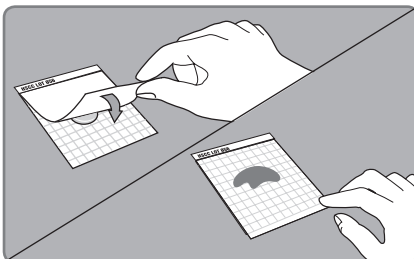
- for acid products, use NaOH 1N,
- for alkaline products, use HCl 1N.

Do not use buffers containing citrate, bisulphite or thiosulphate; they can inhibit growth.

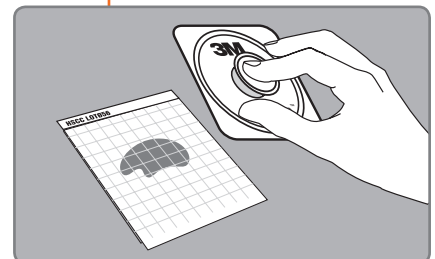
Inoculation



7 Place Petrifilm plate on **level** surface. Lift top film. With pipette **perpendicular** to Petrifilm plate, place **5mL** of sample onto center of bottom film.



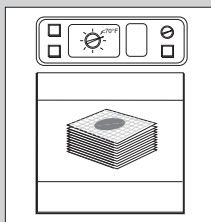
8 **Carefully** roll top film down to avoid trapping air bubbles. Do **not** let top film drop.



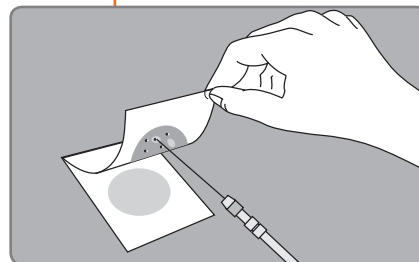
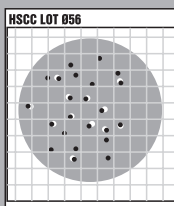
9 Place the **High-Sensitivity plate spreader** on top film over inoculum. Distribute sample with a **gentle pressure** on the handle of the spreader. Do not twist or slide the spreader. Lift spreader. Wait **2 to 5 minutes** for gel to solidify.



Incubation



Interpretation



10 Incubate plates with clear side up in stacks of **up to 10**. Incubation time and temperature vary by method*.

11 Petrifilm plates can be counted with a standard colony counter or other illuminated magnifier. Refer to the Interpretation Guide section when reading results.

12 Colonies may be isolated for further identification. Lift top film and pick the colony from the gel.

Most common methods used in Europe

- AFNOR validated method
3M 01/7-03/99: incubate 24h ± 2h at 30°C ± 1°C or 35°C ± 1°C or 37°C ± 1°C, for total coliforms. Count red colonies with gas.
- incubate 24h ± 2h at 44°C ± 1°C, for thermotolerant coliforms. Incubator humidification is required at this elevated temperature. Count all red colonies.

Most common methods used in the United States

- incubate 24h ± 2h at 32 ± 1°C (dairy products)
- incubate 24h ± 2h at 35 ± 1°C (all foods, except dairy products)

*See product's package insert.

Dilutions

Recommended dilutions

- For cultured dairy products, butter and dry dairy products, a 1:10 dilution is recommended. This yields a 2-coliform-per-gram sensitivity.
- For heavy and light cream, ice cream, chocolate milk, fermented cream, a 1:5 dilution is recommended. This yields a 1-coliform-per-gram sensitivity.
- Raw milk, pasteurized and low fat milk may be plated directly.



Food Safety

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