



Petrifilm®

Interpretation Guide

The Neogen® Petrifilm® Select *E. coli* Count Plate is a sample-ready culture medium system which contains selective agents, nutrients, a cold-water-soluble gelling agent, and an indicator of glucuronidase activity, 5-bromo-4-chloro-3-indolyl-D-glucuronide (BCIG), which facilitates colony enumeration.



SEC

Select *E. coli* Count Plate

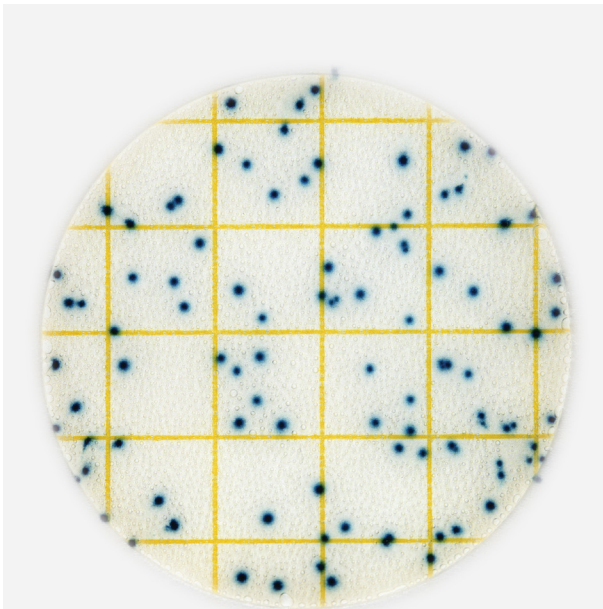


Figure 1

***E. coli* count = 97**

About 97% of *E. coli* strains are thermotolerant and produce beta-glucuronidase, an enzyme that reacts with BCIG indicator in Petrifilm Select *E. coli* Count Plates to produce dark green to blue-green colonies.

Petrifilm Select *E. coli* Count Plates will not detect *E. coli* O157 as most strains are atypical. They are glucuronidase negative, and will not produce a blue-green colony.

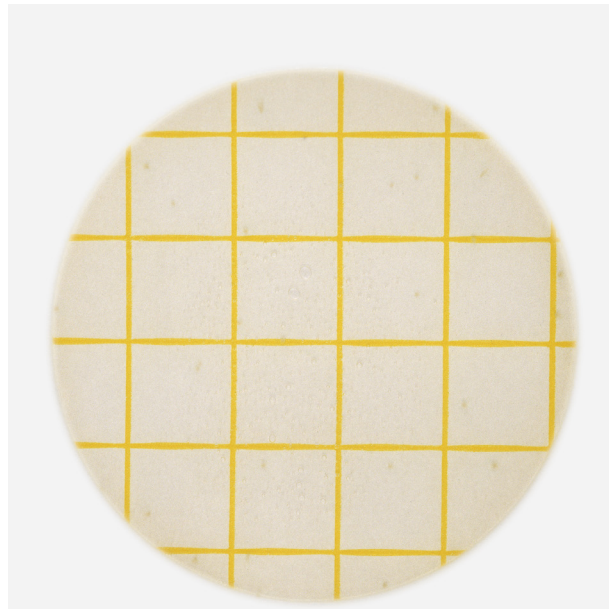


Figure 2

***E. coli* count = 0**

Colonies other than *E. coli* are difficult to see because they are colorless to a light grey-beige.

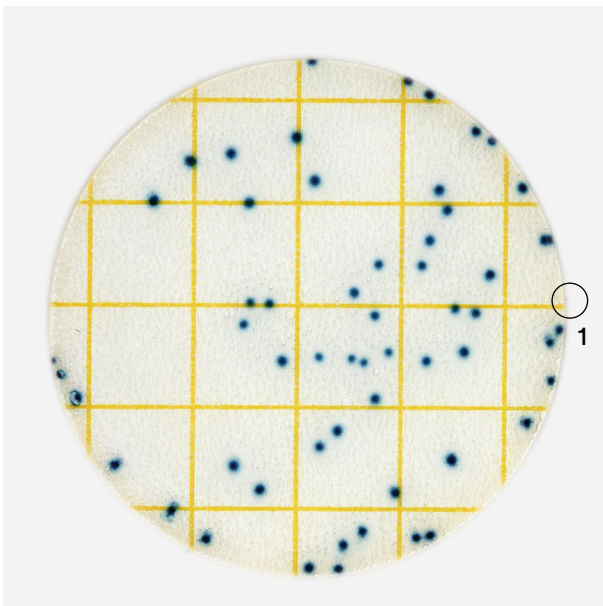


Figure 3

***E. coli* count = 56**

Do not count colonies on the foam dam because they are removed from the selective influence of the medium. See Circle 1.

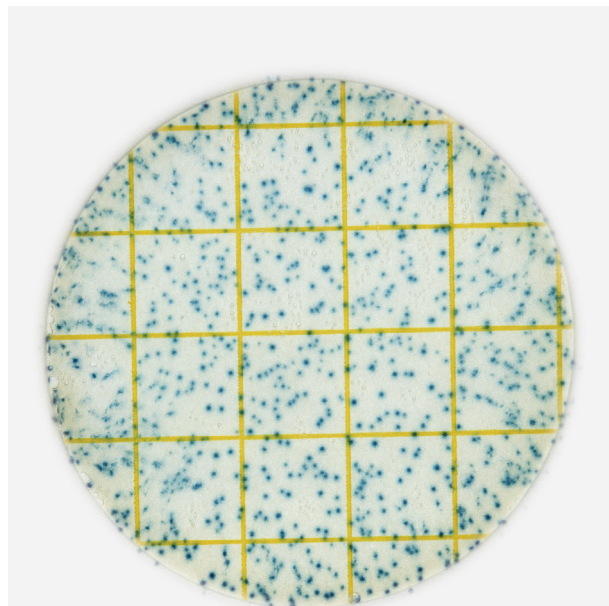


Figure 4

Estimated *E. coli* count = 740

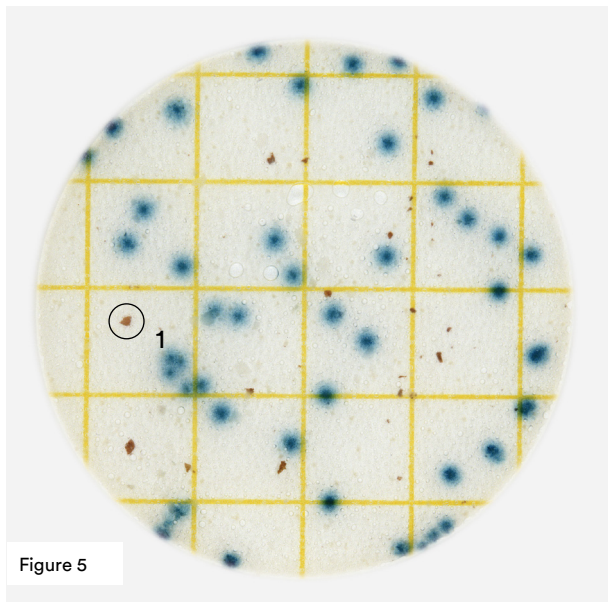
Estimates can be made on plates containing greater than 150 colonies by counting the number of colonies in two or more representative squares and determining the average number per square. Multiply the average number by 20 to determine the estimated count per plate. The circular growth area is approximately 20 cm².

For a more accurate count, further dilution of the sample may be necessary.

Interference from Food Products

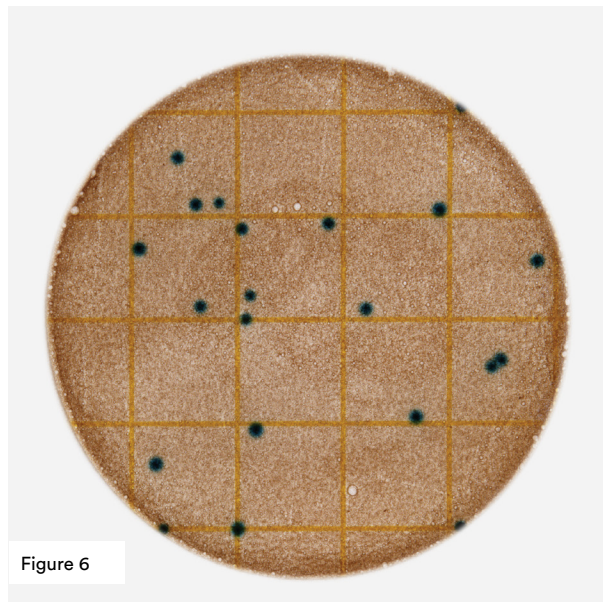
Petrifilm Select *E. coli* Count Plates have been evaluated using samples from many, but not all foods. Foods tested include certain fresh and frozen meats, vegetables and seafood; frozen prepared meals; and fresh, fermented and dry dairy foods. In a limited number of cases, such as liver, the food may interfere with enumeration.

To reduce such food interference, dilute the sample further.



***E. coli* count = 42**

E. coli colonies can easily be distinguished from food particles which are often irregularly shaped and variable in size and color. Circle 1 shows nut particles.



***E. coli* count = 21**

Some dark foods may produce a colored background that makes *E. coli* colonies less distinguishable. Further dilutions will lighten the background color making the *E. coli* colonies easier to count.

Figure 6 shows cocoa powder diluted 1:50.

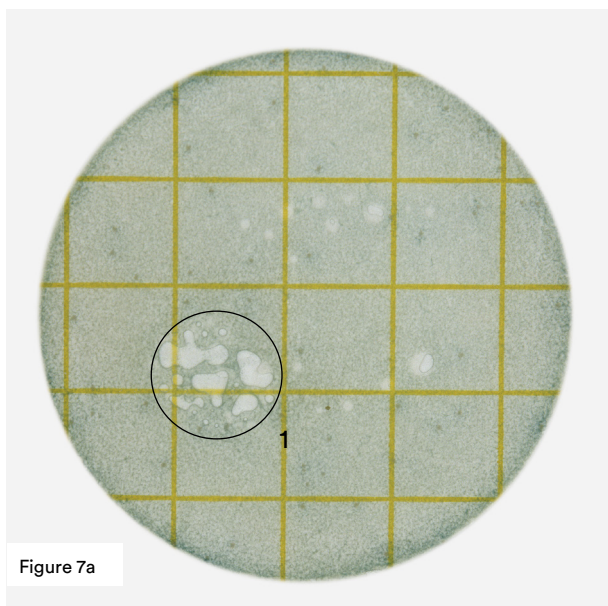


Figure 7a

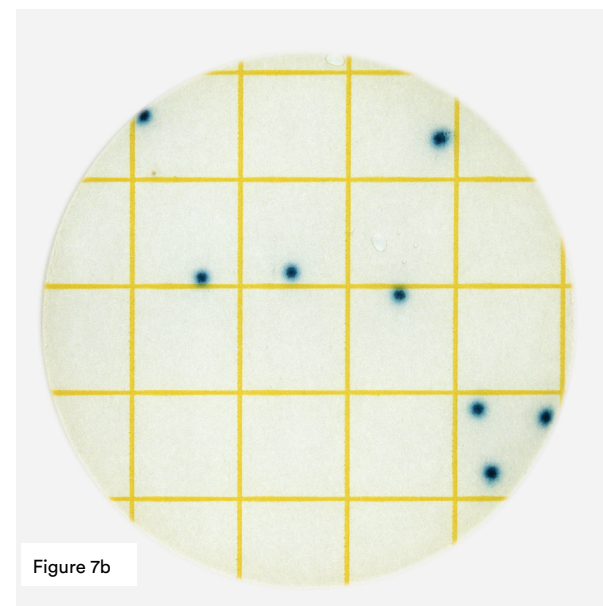


Figure 7b

Raw liver contains β -glucuronidase which produces a blue-green background color on the growth area on the plates, making the *E. coli* colonies less distinguishable. Further dilution will lighten the background color making the *E. coli* colonies easier to count and will help to distinguish food interference from TNTC plates that have confluent colonies (see Figure 13). Artifact bubbles may result from improper inoculation of the plate or from trapped air from the sample. See Circle 1.

Variability in *E. coli* Colony Appearance

Glucuronidase-positive *E. coli* colonies may vary in size, color intensity and shape, depending on the strain itself, the food and the influence of external factors such as testing protocols. The blue-green *E. coli* colonies may have gas bubbles associated with them.

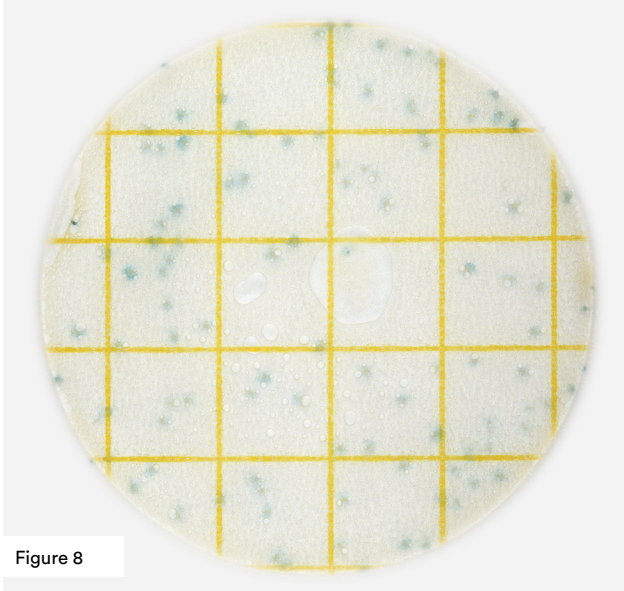


Figure 8

E. coli count = 92

Pale green colonies may result either from *E. coli* that are weak producers of glucuronidase or from an interaction with food such as those containing high acid or high sugar.

Figure 8 shows a highly acidic fermented dairy product.

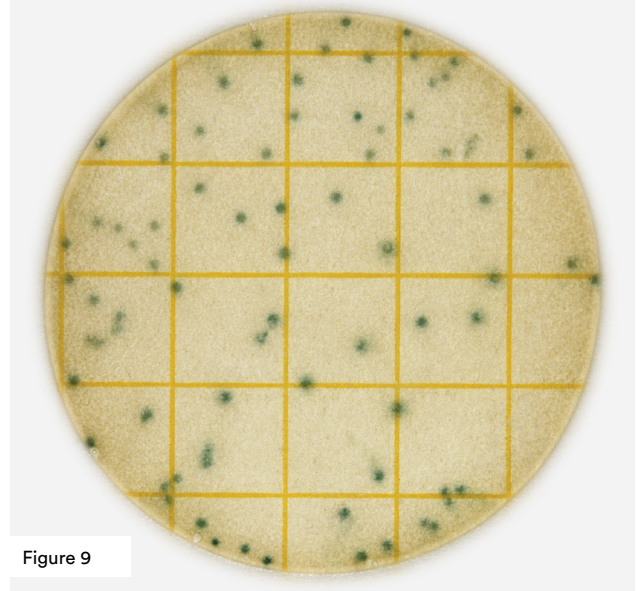


Figure 9

E. coli count = 75

Greenish-brown color variability may appear with some foods.

Figure 9 shows a kidney sample.

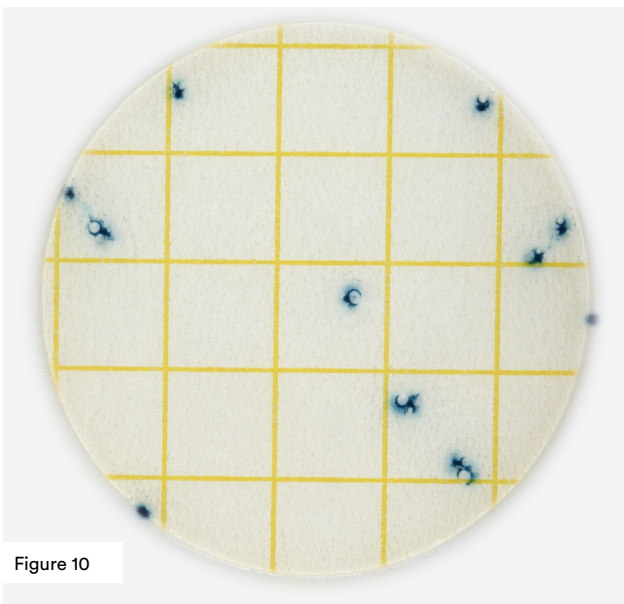


Figure 10

E. coli count = 10

E. coli colonies may have gas bubbles associated with them depending on the *E. coli* strain and the food. Count all colonies with or without gas.

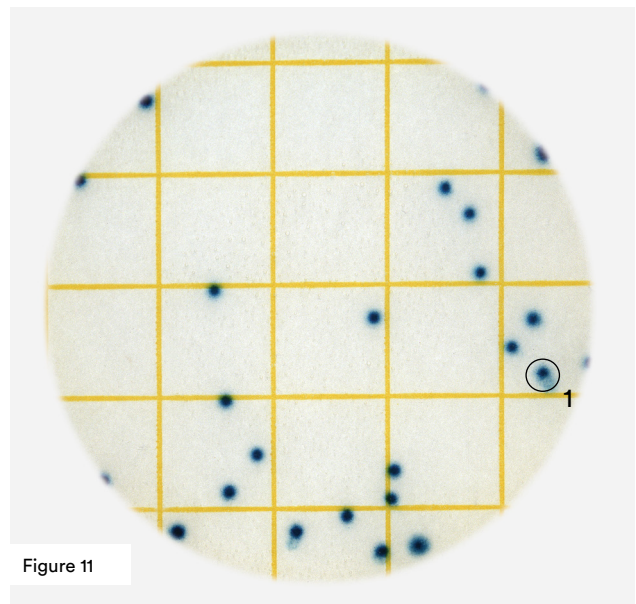


Figure 11

E. coli count = 21

Smeared colonies may appear. See Circle 1.

To minimize the production of smeared colonies spread immediately after inoculation, pressing gently on the center of the spreader.

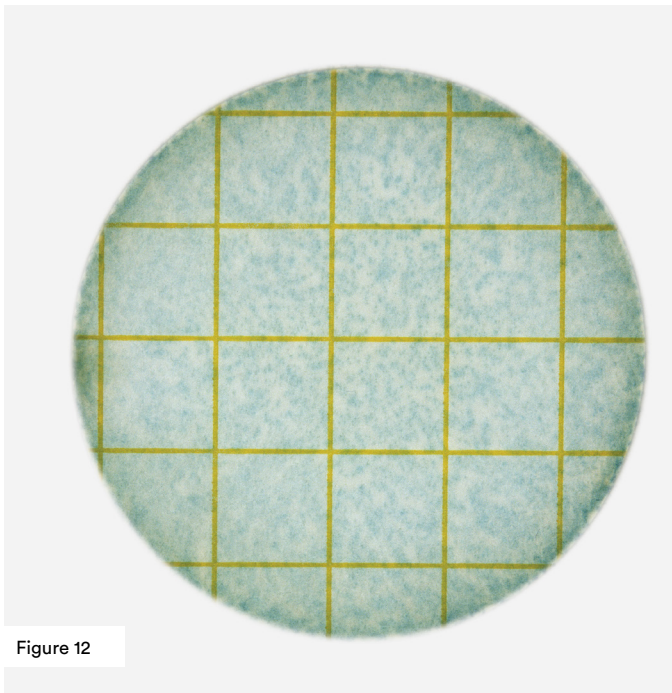


Figure 12

***E. coli* count = Too numerous to count (TNTC)**

When present in large numbers, *E. coli* may appear as small, indistinct colonies.

For a more accurate count, further dilution of the sample may be necessary.

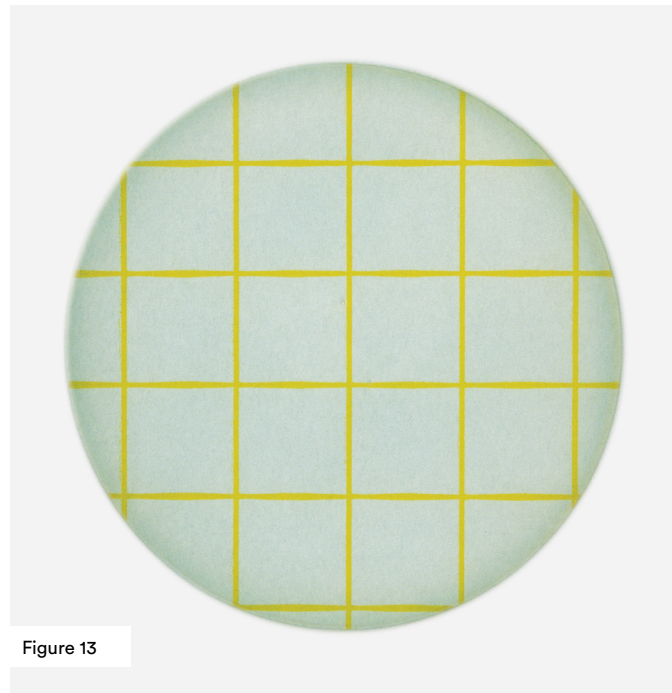


Figure 13

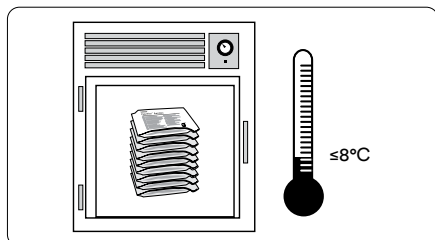
***E. coli* count = TNTC**

High concentrations of *E. coli* will cause the entire growth area to turn blue-green.

For a more accurate count, further dilution of the sample may be necessary.

Reminders For Use

Storage



01

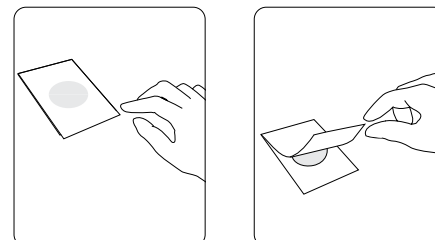
Store unopened packages at ≤8°C (≤46°F). Use before expiration date on package. Just prior to use, allow unopened pouches to come to room temperature before opening.



02

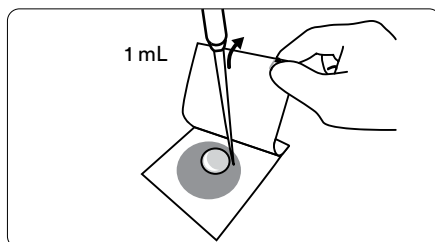
To seal opened package, fold the end of the pouch over and apply adhesive tape. Store resealed pouches in a cool dry place for no longer than four weeks. **To prevent exposure to moisture, do not refrigerate opened pouches.**

Inoculation



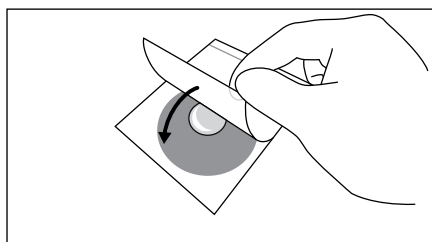
03

Place Petrifilm Select *E. coli* Count Plate on flat, level surface. Lift top film.



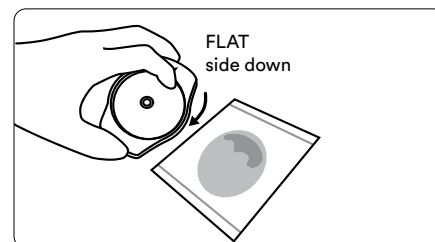
04

With pipette perpendicular to the plate, place 1 mL of sample onto center of bottom film.



05

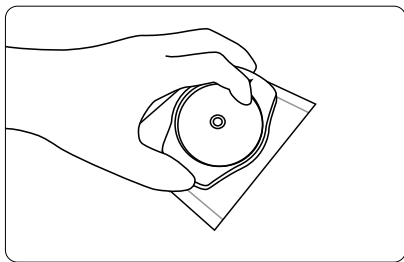
Carefully roll top film down to avoid trapping air bubbles.



06

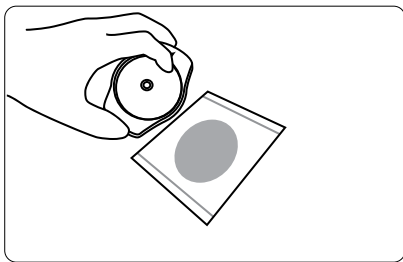
With flat side down, place Petrifilm Spreader on top film over inoculum.

Interpretation



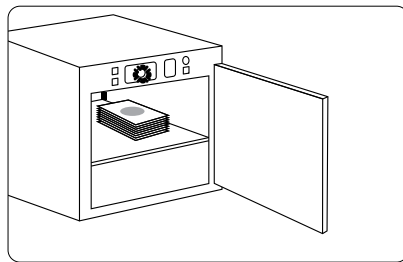
07

Gently apply pressure on spreader to distribute inoculum over circular area. Do not twist or slide the spreader.



08

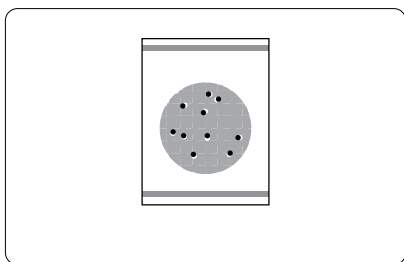
Lift spreader. Wait at least one minute to permit the gel to form.



09

Incubate plates with clear side up in stacks of up to 20. It may be necessary to humidify the incubator to minimize moisture loss.

Please refer to the product instructions for third party validated methods.



10

Petrifilm Select *E. coli* Count Plates can be counted with the Petrifilm Plate Reader Advanced, a standard colony counter or other illuminated magnifier. Colonies may be isolated for further identification. Lift top film and pick the colony from the gel.

Use Appropriate Sterile Diluents

Butterfield's phosphate buffered dilution water, 0.1% peptone water, peptone salt diluent, buffered peptone water, quarter-strength Ringer's solution, dipotassium hydrogen phosphate, saline solution (0.85-0.90%), Wide-Spectrum Neutralizer, bisulfite-free letheen broth or distilled water.

For optimal growth and recovery of the microorganisms, adjust the pH of the sample suspension to 6.5-7.5.

Do not use diluents containing citrate, bisulfite or thiosulfate with Petrifilm Select *E. coli* Count Plates; they can inhibit growth.

If citrate buffer is indicated in the standard procedure, substitute with one of the buffers listed above, warmed to 40-45°C (104-113°F).

Neogen offers a full line of products to accomplish a variety of your microbial testing needs.

For more product information, visit info.neogen.com/petrifilm

User's Responsibilities: Neogen Petrifilm Plate performance has not been evaluated with all combinations of microbial flora, incubation conditions and food matrices. It is the user's responsibility to determine that any test methods and results meet the user's requirements. Should re-printing of this Interpretation Guide be necessary, user's print settings may impact picture and color quality.

For detailed CAUTIONS, DISCLAIMER OF WARRANTIES/LIMITED REMEDY and LIMITATION OF NEOGEN LIABILITY, STORAGE AND DISPOSAL information and INSTRUCTIONS FOR USE, see product instructions.

