3M | Petrifilm™

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KEY diagnostics
T: 02 8212 4074 F: 02 9423 6992
info@keydiagnostics.com.au
www.keydiagnostics.com.au
PO Box 1038, Gymea, NSW, 2227

Product Instructions

- EN Lactic Acid Bacteria Count Plate
- FR Test bactéries lactiques
- DE Milchsäurebakterien Zählplatte
- IT Piastra per il conteggio dei batteri lattici
- ES Placas para Recuento de Bacterias Acido Lácticas
- NL Melkzuurbacterie Telplaat
- SV Lactic Acid Bacteria Count Plate
- DA Mælkesyrebakterie Tælleplade
- NO Petrifilmer for melkesyrebakterier
- FI Maitohappobakteerin Kasvatusalusta
- PT Placa para Contagem de Bactérias Ácido Láticas
- EL Πλακίδιο Καταμέτρησης Βακτηρίων Γαλακτικού Οξέως
- PL Płytka do określania ilości bakterii kwasu mlekowego
- RU Тест-пластина для подсчета молочнокислых бактерий
- TR Laktik Asit Bakterisi Sayım Plakası
- JA 乳酸菌数測定用プレート
- ZH 乳酸菌 测试片
- TH Lactic Acid Bacteria Count Plate
- KO 유산균 측정용 플레이트

LAB
Lactic Acid Bacteria

3M | Petrifilm™

Product Instructions

Lactic Acid Bacteria Count Plate

Product Description and Intended Use

The 3M™ Petrifilm™ Lactic Acid Bacteria Count (LAB) Plate is a self-contained, sample-ready-culture-medium system which contains nutrients, selective agents, a cold-water-soluble gelling agent, and a tetrazolium indicator that facilitates colony enumeration. The 3M Petrifilm LAB Plate contains oxygen scavenging compounds which create an anaerobic environment for the recovery of homofermentative and heterofermentative lactic acid bacteria in the food and beverage industries. Lactic acid bacteria are defined as non-spore forming, Gram positive cocci or rods, which produce lactic acid as a result of carbohydrate fermentation[7]. Homofermentative lactic acid bacteria primarily produce lactic acid whereas heterofermentative lactic acid bacteria produce gas in addition to lactic acid. On the 3M Petrifilm LAB Plate homofermentative lactic acid bacteria appear as red colonies without gas; heterofermentative colonies appear as red colonies with an associated gas bubble.

3M Petrifilm LAB Plate may not detect *Streptococcus thermophilus* which is commonly used as a starter culture in certain fermented products such as yogurt.

The 3M Petrifilm LAB Plate components are decontaminated though not sterilized. 3M Food Safety is certified to International Organization for Standardization (ISO) 9001 for design and manufacturing. The 3M Petrifilm LAB Plates have not been evaluated with all possible food products, food processes, testing protocols or with all possible microorganism strains.

Safety

The user should read, understand, and follow all safety information in the instructions for the 3M Petrifilm LAB Plate. Retain the safety instructions for future reference.

⚠ **WARNING:** Indicates a hazardous situation, which, if not avoided, could result in death or serious injury and/or property damage.

NOTICE: Indicates a potentially hazardous situation which, if not avoided, could result in property damage.

⚠ WARNING

To reduce the risks associated with exposure to biohazards and environmental contamination:

- Follow current industry standards and local regulations for disposal of biohazardous waste.

To reduce the risks associated with release of contaminated product:

- Follow all product storage instruction contained in the product instructions.
- Do not use beyond the expiration date.

To reduce the risks associated with bacterial infection and workplace contamination:

- Perform the 3M Petrifilm LAB Plate testing in a properly equipped laboratory under the control of a skilled microbiologist.
- The user must train its personnel in current proper testing techniques: for example, Good Laboratory Practices[1], ISO/IEC 17025[2] or ISO 7218[3].

To reduce the risks associated with misinterpretation of or inaccurate results:

- 3M has not documented the 3M Petrifilm LAB Plates for use in industries other than food and beverage. For example, 3M has not documented the 3M Petrifilm LAB Plates for testing water, pharmaceuticals, or cosmetics.
- Do not use the 3M Petrifilm LAB Plates in the diagnosis of conditions in humans or animals.
- The 3M Petrifilm LAB Plates do not differentiate any one microorganism strain from another.
- To prevent exposure to moisture, do not refrigerate opened pouches. The freezer that is used for open pouch storage must not have an automatic defrost cycle as this would repeatedly expose the plates to moisture which can damage the plates.
- Do not use 3M Petrifilm LAB Plates that show discoloration.
- Do not use diluents containing citrate or thiosulfate with the 3M Petrifilm LAB Plates; they can inhibit growth.

NOTICE

To avoid inaccurate results and maintain the modified atmosphere:

- Do not lift the top film of the plate unless picking colonies.

Consult the Safety Data Sheet for additional information.

If you have questions about specific applications or procedures, please visit our website at www.3M.com/foodsafety or contact your local 3M representative or distributor.

User Responsibility

Users are responsible for familiarizing themselves with product instructions and information. Visit our website at **www.3M.com/foodsafety**, or contact your local 3M representative or distributor for more information.

When selecting a test method, it is important to recognize that external factors such as sampling methods, testing protocols, sample preparation, handling, and laboratory technique may influence results.

It is the user's responsibility in selecting any test method or product to evaluate a sufficient number of samples with the appropriate matrices and microbial challenges to satisfy the user that the chosen test method meets the user's criteria.

It is also the user's responsibility to determine that any test methods and results meet its customers' and suppliers' requirements.

As with any test method, results obtained from use of any 3M Food Safety product do not constitute a guarantee of the quality of the matrices or processes tested.

Limitation of Warranties / Limited Remedy

EXCEPT AS EXPRESSLY STATED IN A LIMITED WARRANTY SECTION OF INDIVIDUAL PRODUCT PACKAGING, 3M DISCLAIMS ALL EXPRESS AND IMPLIED WARRANTIES, INCLUDING BUT NOT LIMITED TO, ANY WARRANTIES OF MERCHANTABILITY OR FITNESS FOR A PARTICULAR USE. If any 3M Food Safety Product is defective, 3M or its authorized distributor will, at its option, replace or refund the purchase price of the product. These are your exclusive remedies. You must promptly notify 3M within sixty days of discovery of any suspected defects in a product and return it to 3M. Please call Customer Service (1-800-328-1671 in the U.S.) or your official 3M Food Safety representative for a Returned Goods Authorization.

Limitation of 3M Liability

3M 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 3M's liability under any legal theory exceed the purchase price of the product alleged to be defective.

Storage

Store the unopened 3M Petrifilm LAB Plate pouches at frozen or refrigerated temperature equal to -20 to 8°C (-4 to 46°F). Just prior to use, allow unopened pouches to come to room temperature equal to 20 to 25°C (68 to 77°F) and <60% RH before opening. Return the unused 3M Petrifilm LAB Plates to pouch. Seal by folding the end of the pouch over and applying adhesive tape. **To prevent exposure to moisture, do not refrigerate opened pouches.** Store resealed pouches in a cool dry place for no longer than four weeks. It is recommended that resealed pouches of the 3M Petrifilm LAB Plates be stored in a freezer equal to or less than -15°C (5°F) for no longer than four weeks if the laboratory temperature exceeds 25°C (77°F) and/or the laboratory is located in a region where the relative humidity exceeds 50% (with the exception of air-conditioned premises).

To store resealed pouches in a freezer, place 3M Petrifilm LAB Plates in a sealable container. To remove the frozen 3M Petrifilm LAB Plates for use, open the container, remove the plates that are needed and immediately return remaining plates to the freezer in the sealed container. The freezer that is used for open pouch storage must not have an automatic defrost cycle as this would repeatedly expose the plates to moisture which can damage the plates.

Do not use 3M Petrifilm LAB Plates that show discoloration. Expiration date and lot number are noted on each package of the 3M Petrifilm LAB Plates. The lot number is also noted on individual 3M Petrifilm LAB Plates.

⚠Disposal

After use, the 3M Petrifilm LAB Plates may contain microorganisms that may be a potential biohazard. Follow current local, regional, national and industry standards for disposal.

Instructions for Use

Follow all instructions carefully. Failure to do so may lead to inaccurate results.

Sample Preparation

1. Use appropriate sterile diluents:

   Butterfield's phosphate-buffered dilution water, buffered peptone water, 0.1% peptone water, saline (0.85-0.90%), letheen broth, modified letheen broth or peptone salt diluent (Maximum Recovery Diluent).

   Do not use diluents containing citrate or thiosulfate with the 3M Petrifilm LAB Plates; they can inhibit growth.

2. Blend or homogenize the sample.

Plating

1. Place the 3M Petrifilm LAB Plate on a flat, level surface.
2. Lift the top film and with the pipette perpendicular to the inoculation area dispense 1 mL of sample suspension onto the center of bottom film.

3. Roll the top film down onto the sample to prevent trapping air bubbles.
4. Place the 3M™ Petrifilm™ Flat Spreader (catalog #6425) on the center of the 3M Petrifilm LAB Plate. Press gently on the center of the 3M Petrifilm Flat Spreader to distribute the sample evenly. Spread the inoculum over the entire 3M Petrifilm LAB Plate growth area before the gel is formed. Do not slide the 3M Petrifilm Flat Spreader across the film.
5. Remove the 3M Petrifilm Flat Spreader and leave the 3M Petrifilm LAB Plate undisturbed for at least one minute to permit the gel to form.

Incubation

Incubate the 3M Petrifilm LAB Plates in a horizontal position with the clear side up in stacks of no more than 20.

Incubate the 3M Petrifilm LAB Plates for 48 hours ± 3 hours at 28 to 37°C. Several incubation times and temperatures can be used depending on current local reference methods, some of which are listed in the "**Specific Instructions for Validated Methods**" section.

Interpretation

1. The 3M Petrifilm LAB Plates can be counted using a standard colony counter or other illuminated magnifier. Count all red colonies regardless of size or intensity. Do not count colonies on the dam since they are removed from the selective influence of the medium. Do not count artifact bubbles that may be present.
2. For total lactic acid bacteria counts, count all red colonies with or without gas.
3. Heterofermentative lactic acid bacteria are defined as colonies that are red and closely associated (within one colony diameter) with entrapped gas. Red colonies without gas are defined as homofermentative lactic acid bacteria.
4. The circular growth area is approximately 30 cm^2. The counting range for 3M Petrifilm LAB Plate is lower than or equal to 150 red colonies with gas and/or lower than or equal to 300 red colonies without gas. Estimates can be made on the 3M Petrifilm LAB Plates containing greater than 150 or 300 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 30 to determine the estimated count per plate.
5. 3M Petrifilm LAB Plates with colony counts too numerous to count (TNTC) may have one or more of the following characteristics: many small colonies, many gas bubbles, and a deepening of the gel color from blue to pink-purple. High concentrations of colonies on the 3M Petrifilm LAB Plates will cause the entire growth area to become deep-blue to purple with a pink halo around the outer edge of the plate. Occasionally, distribution of colonies or gas bubbles may appear irregular. When any of these occur, record results as TNTC. When an actual count is required, plate at a higher dilution.
6. Where necessary, colonies may be isolated for further identification. Lift the top film and using proper technique, pick the colony from the gel. Test using standard procedures.
7. If the 3M Petrifilm LAB Plates cannot be counted immediately after removal from the incubator, they may be stored for later enumeration by freezing in a sealable container at temperatures lower than or equal to -15°C (5°F) for no longer than one week.

For further information refer to the "3M™ Petrifilm™ LAB Plate Interpretation Guide." If you have questions about specific applications or procedures, please visit our website at www.3M.com/foodsafety or contact your local 3M representative or distributor.

Specific Instructions for Validated Methods

AOAC® *Performance Tested Method*℠ Certificate #041701

In an AOAC RI PTM study, the 3M Petrifilm LAB Count Plate method was found to be equivalent to the average log counts of Compendium of Methods for the Microbiological Examination of Foods (CMMEF) Chapter 19, Fifth Edition and the ISO 15214: *Microbiology of food and animal feeding stuffs – Horizontal method for the enumeration of mesophilic lactic acid bacteria –colony-count technique at 30°C,* First edition, 1998-08-01.

Scope of Validation: Cold smoked salmon, cream pastry, creamy salad dressing, deli chicken, deli ham, deli turkey, duck pate, pickled herring, kimchi, mayonnaise, mustard potato salad, terrines, yogurt, chicken sausage, pepperoni, cottage cheese, ready-to-bake pizza, and stainless steel (environmental surface).

Incubation:

Incubate the 3M Petrifilm LAB Plates 48 hours ± 3 hours at 28°C ± 1°C to 37°C ± 1°C.

PERFORMANCE TESTED AOAC RESEARCH INSTITUTE LICENSE NUMBER 041701

NF VALIDATION by AFNOR Certification

NF VALIDATION certified method in compliance with ISO 16140-2[8] in comparison to ISO 15214[6]

Use the following details when implementing the above Instructions for Use:

Scope of the validation:

All human foods (excluding yogurts) and industrial environmental samples.

Sample preparation:

Use only ISO listed diluents[5] or letheen broth after sanitation.

Incubation:

Incubate 3M Petrifilm LAB Plates 48 hours ± 3 hours at 30°C ± 1°C.

Interpretation:

Calculate the number of microorganisms present in the test sample according to ISO 7218[3] for one plate per dilution. Estimates are outside the scope of the NF Validation certification (cf Interpretation paragraph 4).

3M 01/19-11/17
ALTERNATIVE ANALYTICAL METHODS FOR AGRIBUSINESS
http://nf-validation.afnor.org/en

For more information about end of validity, please refer to NF VALIDATION certificate available on the website mentioned above.

References

1. U.S. Food and Drug Administration. Code of Federal Regulations, Title 21, Part 58. Good Laboratory Practice for Nonclinical Laboratory Studies.
2. ISO/IEC 17025, General requirements for the competence of testing and calibration laboratories.
3. ISO 7218, Microbiology of food and animal feeding stuffs – General requirements and guidance for microbiological examinations.
4. FDA Bacteriological Analytical Manual (BAM), Reagents Index for BAM found at: http://www.fda.gov/Food/FoodScienceResearch/LaboratoryMethods/ucm055791.htm.
5. ISO 6887, Microbiology of food and animal feeding stuffs – Preparation of test samples, initial suspension and decimal dilutions for microbiological examination.
6. ISO 15214, Microbiology of food and animal feeding stuffs – Horizontal method for the enumeration of mesophilic lactic acid bacteria – Colony-count technique at 30 degrees C.
7. American Public Health Association. Compendium of Methods for the Microbiological Examination of Foods 5th Edition.
8. ISO 16140-2, Microbiology of the food chain - Method Validation - Protocol for the validation of alternative (proprietary) methods against a reference method.

Explanation of Symbols

www.3M.com/foodsafety/symbols

3M Food Safety

3M United States
3M Center
Bldg. 275-5W-05
St. Paul, MN 55144-1000
USA
1-800-328-6553

3M Canada
Post Office Box 5757
London, Ontario N6A 4T1
Canada
1-800-563-2921

3M Latin America
3M Center
Bldg. 275-5W-05
St. Paul, MN 55144-1000
USA
1-954-340-8263

3M Europe and MEA
3M Deutschland GmbH
Carl-Schurz-Strasse 1
D41453 Neuss/Germany
+49-2131-14-3000

3M United Kingdom PLC
Morley Street, Loughborough
Leicestershire
LE11 1EP
United Kingdom
+(44) 1509 611 611

3M Österreich GmbH
Euro Plaza
Gebaude J, A-1120 Wien
Kranichberggasse 4
Austria
+(43) 1 86 686-0

3M Asia Pacific
No 1, Yishun Avenue 7
Singapore, 768923
65-64508869

3M Japan
3M Health Care Limited
6-7-29, Kita-Shinagawa
Shinagawa-ku, Tokyo
141-8684 Japan
81-570-011-321

3M Australia
Bldg A, 1 Rivett Road
North Ryde, NSW 2113
Australia
61 1300 363 878

3M Health Care
2510 Conway Ave
St. Paul, MN 55144 USA
www.3M.com/foodsafety

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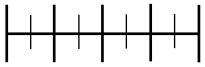
Scale:  1 Inch

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