

Product Instructions

Yeast and Mold Count Plate





Product Instructions

Aqua | Yeast and Mold Count Plate

Description

The 3M™ Petrifilm™ Aqua Yeast and Mold Count (AQYM) Plate is a sample-ready culture medium system which contains nutrients supplemented with antibiotics, a cold-water-soluble gelling agent, and an indicator that facilitates yeast and mold enumeration. Petrifilm AQYM Plates are used for the enumeration of yeast and mold in bottled water industries. Petrifilm AQYM Plate components are decontaminated though not sterilized. 3M Food Safety is certified to ISO (International Organization for Standardization) 9001 for design and manufacturing.

⚠ Cautions

3M has not documented Petrifilm AQYM Plates for use in industries other than bottled water. For example, 3M has not documented Petrifilm AQYM Plates for testing surface and municipal waters, or waters used in the pharmaceutical or cosmetic industries. The use of Petrifilm AQYM Plates to test water samples in compliance with local water testing regulations is at the sole discretion and responsibility of the end-user.

Petrifilm AQYM Plates have not been tested with all possible bottled water samples, testing protocols or with all possible strains of yeast and mold.

Do not use Petrifilm AQYM Plates in the diagnosis of conditions in humans or animals.

User Responsibility

No one culture medium will always recover the exact same strains or enumerate a particular strain exactly as does another medium. In addition, external factors such as sampling methods, testing protocols, preparation time and handling may influence recovery and enumeration.

It is the user's responsibility in selecting any test method to evaluate a sufficient number of bottled water samples 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' or suppliers' requirements.

As with any culture medium, Petrifilm AQYM Plate results do not constitute a guarantee of quality of bottled water products that are tested with the plates.

⚠ The user must train its personnel in current proper testing techniques: for example, Good Laboratory Practices¹ or ISO 17025².

Disclaimer of Warranties / Limited Remedy

UNLESS OTHERWISE PROHIBITED BY LAW, 3M DISCLAIMS ALL EXPRESS AND IMPLIED WARRANTIES, INCLUDING, BUT NOT LIMITED TO, ANY WARRANTIES OF MERCHANTABILITY OR FITNESS FOR A PARTICULAR USE. If any Petrifilm Plate is proven to be defective, 3M or its authorized distributor will replace or, at its option, refund the purchase price of any plate. These are your exclusive remedies. You must promptly notify 3M within sixty days of discovery of any suspected defect in a product and return the product to 3M.

Limitation of 3M Liability

UNLESS OTHERWISE PROHIBITED BY LAW, 3M WILL NOT BE LIABLE TO USER OR OTHERS FOR ANY LOSS OR DAMAGE, WHETHER DIRECT, INDIRECT, SPECIAL, INCIDENTAL OR CONSEQUENTIAL DAMAGES, INCLUDING, BUT NOT LIMITED TO, LOST PROFITS. Except where prohibited by law, in no event shall 3M's liability under any legal theory exceed the purchase price of the plates alleged to be defective. Customer may have additional rights and should seek advice in country of purchase.

Storage and Disposal

Store **unopened** Petrifilm Plate pouches refrigerated or frozen at temperatures less than or equal to 8°C (46°F). Just prior to use, allow unopened pouches to come to room temperature before opening. Return unused 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 one month. It is recommended that resealed pouches of Petrifilm Plates be stored in a freezer (see below) 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 opened pouches in a freezer, place Petrifilm Plates in a sealable container. To remove frozen Petrifilm Plates for use, open the container, remove the plates that are needed and immediately return remaining plates to the freezer in the sealed container. Plates should not be used past their expiration date. Do not store open pouches in a freezer with an automatic defrost cycle, as this could damage the plates due to repeated exposure to moisture.

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

⚠ After use, Petrifilm AQYM Plates may contain microorganisms that may be a potential biohazard. Follow current industry standards for disposal.

Instructions for Use

Water Filtration and Plate Incubation

1. Following standard procedures for water analysis, membrane filter water sample using a 47 mm, 0.45 micron pore size Mixed Cellulose Ester (MCE) filter.
2. Carefully lift the top film of the Petrifilm AQYM Plate. Avoid touching the circular growth area. Place the filter in the center of the plate (Figure A).
3. Hydrate the Petrifilm AQYM Plate by placing 1 mL appropriate sterile, hydration diluent in the center of the filter (Figure B). Appropriate sterile hydration diluents include distilled water, deionized (DI) water and reverse-osmosis (RO) water.
4. Slowly roll the top film onto the filter. Minimize trapping air bubbles and creating gaps between the filter and the Petrifilm AQYM Plate. Lightly apply pressure by using the Petrifilm Plate AQYM spreader (Figure C1 and Figure C2).
5. Incubate Petrifilm AQYM Plates at 20-25°C for 3-5 days³ in a horizontal position with the clear side up in stacks of no more than 20.

Interpretation

1. Petrifilm AQYM Plates can be counted using a standard colony counter or other illuminated magnifier.
2. To differentiate yeast and mold colonies on the Petrifilm AQYM, look for one or more of the following characteristics (Figure D, Figure E and Figure F):

YEAST	MOLD
Small colonies	Large colonies
Colonies have defined edges	Colonies have diffuse edges
Pink-tan to blue-green in color	Variable color
Colonies appear raised (3 dimensional)	Colonies appear flat
Colonies have uniform color	Colonies have a dark center*

* Mold colonies on the surface of a filter may not exhibit a dark center.

3. Read final yeast and mold results on day 3-5 depending on the fungi present³.

Note: During the hydration of the Petrifilm AQYM Plate, some colonies may be eluted off of the filter onto the surrounding inoculation area (Figure F and Figure G). Count all colonies on both the filter and the surrounding media.

4. If the plate cannot be counted due to high numbers of colonies or confluent growth, plate at a higher dilution.
5. Petrifilm AQYM Plates use a phosphatase indicator to help detect yeast and mold. Natural phosphatase in water samples may create a uniform blue background color or intense, pinpoint blue spots. Check plates after 24 - 48 hours incubation and note any color present; if color intensity does not change by day 5, the color may be from the phosphatase reaction.

6. Colonies may be isolated for further identification. Lift the top film and pick the colony from the gel or the filter surface (Figure G). When lifting the top film, the filter may adhere to either the top film or the bottom film. If the filter adheres to the top film, separate the filter from the top film and pick colonies. Test using standard procedures.

Note: Delayed counting of Petrifilm AQYM Plates with filters is not recommended.

For further information refer to the appropriate Petrifilm Plate “Interpretation Guide.”

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. American Public Health Association. 1998. Standard Methods for the Examination of Water and Wastewater, 20th Ed. Method 9610D.

Refer to the current versions of the standard methods listed above.



Petrifilm™

Environmental Monitoring Procedures

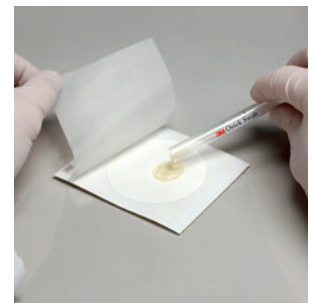
3M™ Petrifilm™ Plates are a convenient and reliable way to detect environmental microbial contamination. The construction of 3M Petrifilm Plates allows them to be used for direct contact or swab contact monitoring procedures, as well as air sampling procedures.



Air Sampling



Surface Contact



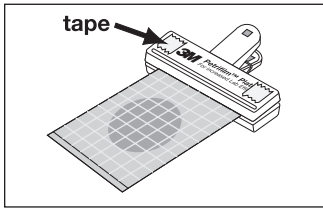
Swabbing

Hydration Procedures for Air or Direct Contact Methods

3M Petrifilm Plate	Hydration*	Storage
Aerobic Count Coliform Count <i>E. coli</i> /Coliform Count Rapid Coliform Count <i>Enterobacteriaceae</i> Count Yeast and Mold Count Rapid Yeast and Mold Count Rapid <i>E. coli</i> /Coliform Count	Hydrate plates with 1 mL of appropriate sterile diluent for a minimum of 1 hour before use. Allow hydrated plates to remain closed for a minimum of 1 hour before use.	Store all hydrated 3M Petrifilm Plates in sealed pouch or plastic bag. Protect plates from light and refrigerate at 2–8°C (36–46°F). Hydrated 3M Petrifilm Aerobic Count Plates may be refrigerated up to 14 days, 3M Petrifilm Rapid Yeast and Mold Count Plates may be refrigerated up to 1 day (24 hours) and all other hydrated 3M Petrifilm Plates may be refrigerated up to 7 days.
Staph Express System	Hydrate plates with 1 mL of appropriate sterile diluent. Refrigerate hydrated plates at 2–8°C (36–36°F) for a minimum of 3 hours before use.	
Rapid Aerobic Count	Hydrate plates with 1 mL of appropriate sterile diluent. For air sampling, refrigerate at 2–8°C (36–46°F) for a minimum of 1 day (24 hours) before use. For direct contact samples, refrigerate at 2–8°C (36–46°F) for a minimum of 3 days before use.	

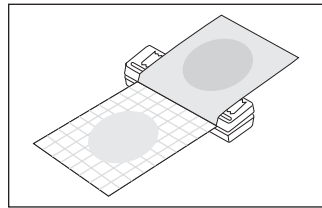
*See relevant 3M Petrifilm Plate product instructions for details and listing of appropriate diluents. If sanitizers are present, use letheen broth for both the direct contact and swab contact methods.

3M Petrifilm Plate Air Sampling Method

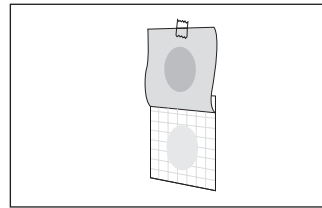


- 1 Use a 3M Petrifilm Plate clip in combination with double-sided tape. Position hinged edge of hydrated 3M Petrifilm Plate into clip. Apply a small piece of double-sided tape to each end of the clip handle.

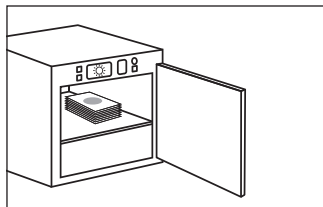
Double-sided tape can also be used with or without clip for positioning of 3M Petrifilm Plates for air sampling.



- 2 **Without touching circular growth area,** lift top film portion of hydrated plate and peel back until outer portion of film adheres to the tape. Make sure top film lies flat across clip.



- 3 Expose 3M Petrifilm Plate to air for no longer than 15 minutes. Remove tape and rejoin the top and bottom films.



- 4 Incubate and enumerate as directed in product instructions. Refer to 3M Petrifilm Plate Interpretation Guide when enumerating results.

Air Sampling Method Results

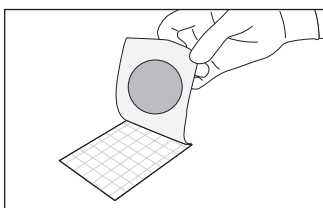
3M Petrifilm Plates: Aerobic Count, Coliform Count, *E. coli*/Coliform Count, Rapid Coliform Count, *Enterobacteriaceae* Count

Results: count/40 cm²

3M Petrifilm Plates: Staph Express Count, Yeast and Mold Count, Rapid Yeast and Mold Count, Rapid Aerobic Count, Rapid *E. coli*/Coliform Count

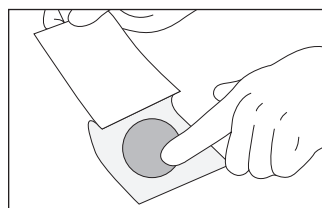
Results: count/60 cm²

3M Petrifilm Plate Direct Contact Method

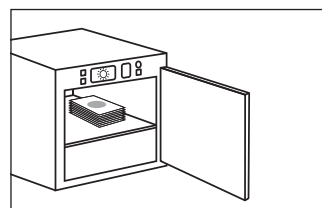


- 1 Using a hydrated 3M Petrifilm Plate, carefully lift top film. Avoid touching circular growth area. Gel will adhere to top film.

3M™ Petrifilm™ Yeast and Mold Count Plates: On occasion, the gel may split (adhering to both the top and bottom films) when the top film is lifted. If this happens, the plate with gel splitting may still be used for air testing, but is not recommended for direct contact use.



- 2 Allow the circular gel portion of the top film to contact the surface being tested. Gently rub fingers parallel to the surface over the outer film side of the gelled area to ensure good contact with surface. Rejoin the top and bottom films.



- 3 Incubate and enumerate as directed in product instructions. Refer to 3M Petrifilm Plate Interpretation Guide when enumerating results.

Direct Contact Method Results

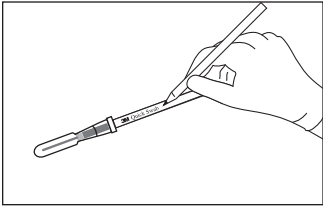
3M Petrifilm Plates: Aerobic Count, Coliform Count, *E. coli*/Coliform Count, Rapid Coliform Count, *Enterobacteriaceae* Count

Results: count/20 cm²

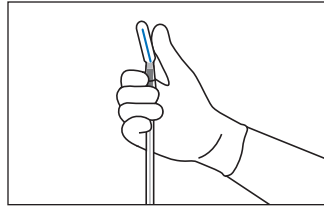
3M Petrifilm Plates: Staph Express Count, Yeast and Mold Count, Rapid Yeast and Mold Count, Rapid Aerobic Count, Rapid *E. coli*/Coliform Count

Results: count/30 cm²

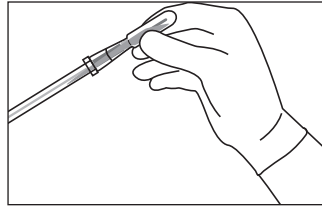
3M™ Quick Swab Method (wet swabbing method)*



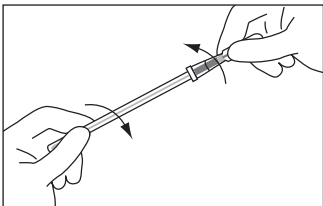
1 Remove the desired quantity of 3M Quick Swabs from the resealable plastic bag. Label the swab.



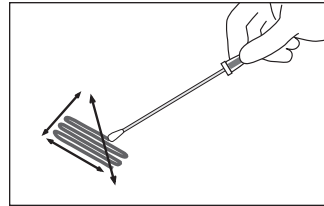
2 At the sampling location, prepare the swab by holding it with the bulb end near your thumb. Bend the red snap valve at a 45° angle until you hear the valve break. This allows the letheen broth to flow into the tube and wet the swab head.



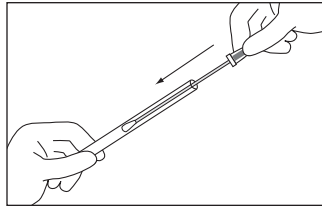
3 Squeeze the bulb of the swab to transfer all of the letheen broth to the tube end of the swab.



4 Twist and pull apart the bulb end of the swab from the tube end of the swab which contains the letheen broth.



5 Hold the swab handle to make a 30° angle with the surface. Firmly rub the swab head slowly and thoroughly over the desired surface area. Rub the head of the swab three times over the surface, reversing direction between alternating strokes.



6 After sampling is complete, securely insert the swab head back into the swab tube and transport to the lab for plating. Plate the letheen broth swab solution as soon as possible.

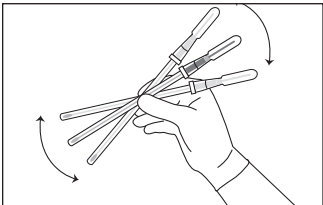
Alternative Swab Method

3M Petrifilm Plates can be used with other swabbing techniques, however the rinse solution used must be compatible with 3M Petrifilm Plates.

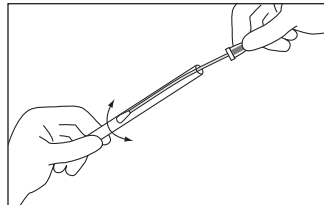
*For 3M Quick Swab dry swabbing method, see 3M Quick Swab product instructions.

Inoculation Procedures

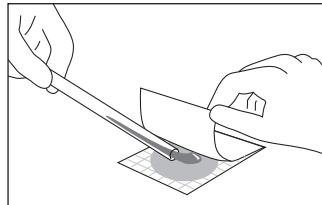
1 mL Inoculation Procedure



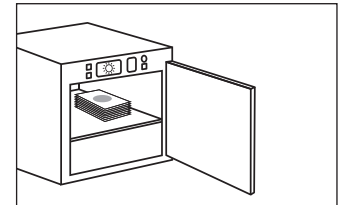
7a In the lab, vigorously shake or vortex the swab for 10 seconds, to release bacteria from the swab tip.



8a Release the contents of the swab tip by pressing and twisting the swab against the wall of the tube.

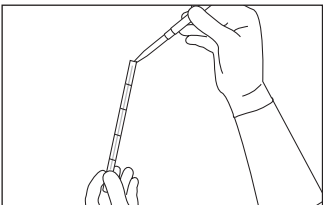


9a Carefully pour entire contents of the tube onto a 1 mL 3M Petrifilm Plate. Follow current industry standards for disposal.

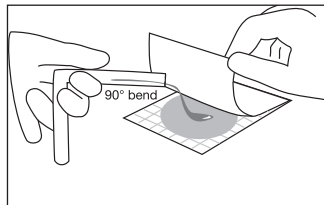


10a Incubate and enumerate as directed in product instructions. Refer to 3M Petrifilm Plate Interpretation Guide when enumerating results.

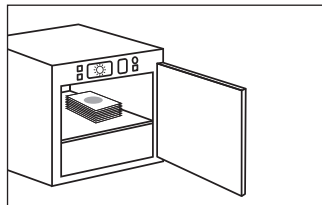
Multi-mL Inoculation Procedure



7b Remove the swab from the tube. Add 1–3 mL of sterile diluent to the swab tube. Replace the swab in the tube. Complete steps 7a and 8a of the 1 mL Inoculation Procedure from above.



8b Use your thumb to bend the swab tube at a 90° angle at the highest mark that has diluent above it. Pour off a 1 mL aliquot onto a 3M Petrifilm Plate. Repeat onto a new plate until the entire sample is used.



9b Incubate and enumerate as directed in product instructions. Refer to 3M Petrifilm Plate Interpretation Guide when reading results.

3M Quick Swab Method Results

1 mL Inoculation Procedures:

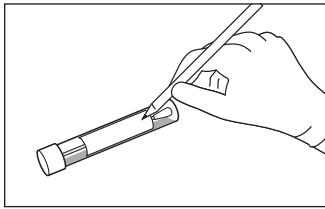
3M Petrifilm Plate count x volume of diluent (1 mL) = total count/area sampled.

Example: If area tested was 5cm² and number of colonies on plate after incubation was 100, your result would be: 100 CFU x 1 mL = 100 CFU/5 cm²

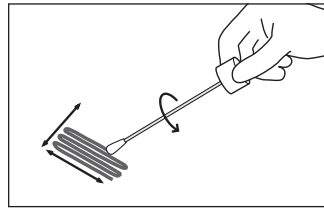
Multi-mL Inoculation Procedures: 3M Petrifilm Plate count x volume of diluent (1 mL + added) = total count/area sampled.

Example: If area tested was 5cm² and 2 mL were added (for total of 3 mL) and number of colonies after incubation was 100, your result would be: 100 CFU x 3 mL = 300 CFU/5 cm²

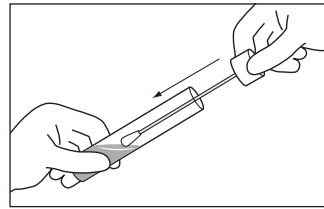
3M™ Swab Sampler Method



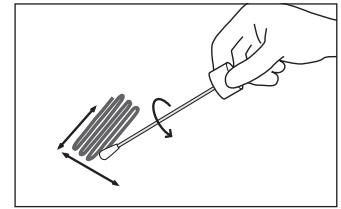
1 Label the 3M Swab Sampler. Unscrew the cap from the tube and aseptically remove the swab from the tube.



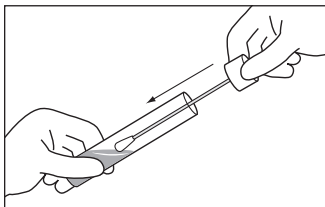
2 Aseptically swab across the sampling surface while rotating the swab.



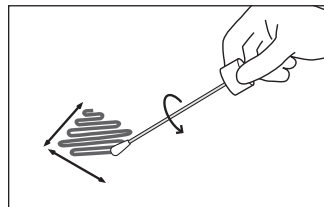
3 Return swab to the tube.



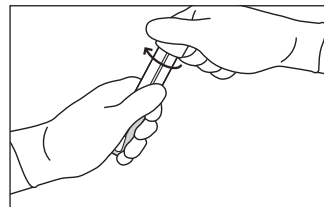
4 Repeat Step 2. Change direction 90° and aseptically swab the surface while rotating the swab.



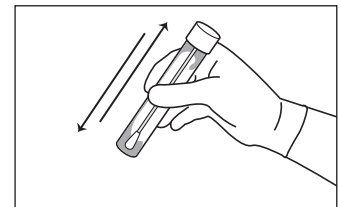
5 Return swab to the tube.



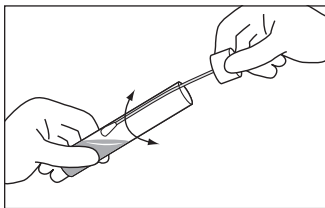
6 Repeat Step 2. Change direction 45° and aseptically swab the same sampling surface while rotating the swab.



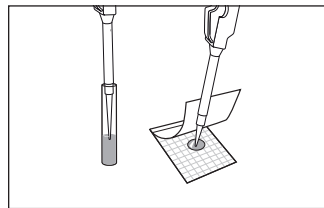
7 Return swab to the tube. Screw cap tight to close.



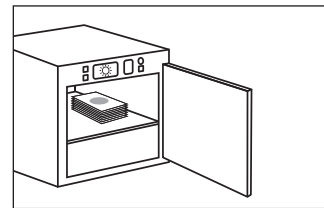
8 In the lab, vigorously shake or vortex the swab for 10 seconds, to release bacteria from the swab tip.



9 Unscrew the cap, release out the contents of the swab tip by pressing and twisting the swab against the wall of the tube. Remove swab from tube.



10 Using a pipettor with a sterile tip, draw 1 mL from the tube and dispense onto a 3M Petrifilm Plate. Repeat for additional plates as needed.



11 Incubate and enumerate as directed in product instructions. Refer to 3M Petrifilm Plate Interpretation Guide when reading results.

3M Swab Sampler Results

3M Petrifilm Plate count x volume of 3M Swab Sampler = total count/area sampled.

Example: If area tested was 5 cm² and a 4 mL 3M Swab Sampler was used and number of colonies on plate after incubation was 100, your result would be: 100 CFU x 4 mL = 400 CFU/5 cm²

Lethen broth 3M Swab Samplers are available in variety of sizes: 1 mL, 4 mL, 5 mL, 10 mL