



PRODUCT SPECIFICATION SHEET

Milk Agar w/ Cetrимide (Twin Pack) (DM349)

Intended Use

Milk Agar w/ Cetrимide (Twin Pack) (DM349) is recommended for detection and enumeration of *Pseudomonas aeruginosa* in water.

Product Summary and Explanation

Microbiological contamination of the swimming pool water can result in pathogenic microorganisms causing infections to bathers. These contaminants can be introduced into pool water from bathers, from the pool filters or occasionally from defects in pool engineering (e.g. that allow the water to be contaminated with sewage). Swimming pool water is usually chlorinated potable water but it can also be from thermal springs or salt water. As *P. aeruginosa* can survive for longer time in water compared to other microorganisms, it is one of the major indicator organisms in the swimming pool. *Pseudomonas aeruginosa* can grow within untreated waters and in biofilms. It can cause skin, ear and eye infections when present in large numbers and outbreaks of skin infections have been linked to swimming pools and spa pools.

Brown and Scott⁽¹⁾ modified Milk Agar for the confirmation of *Pseudomonas aeruginosa* in swimming pool waters. In accordance with ISO Committee under the specifications ISO 8360-1: 1988, Milk Agar with Cetrимide is formulated for the detection and enumeration of *P. aeruginosa* from water.⁽²⁾ *P. aeruginosa* is the only species of *Pseudomonas* or gram-negative rod known to excrete pyocyanin and this pigment production is one of the major identification tool for strains of *P. aeruginosa*.

Principles of the Procedure

Milk Agar w/ Cetrимide contains skim milk powder, milk, peptic digest of animal tissue and yeast extract provide all the necessary nutrients mainly nitrogenous for the growth and metabolism of *P. aeruginosa*. Sodium chloride helps to maintain the osmotic balance of the medium. Cetrимide is a quaternary ammonium, cationic detergent compound, which is inhibitory to a wide variety of bacterial species including *Pseudomonas* species other than *P. aeruginosa*.

Formula / Liter

Ingredients	Gms / Liter
Part A	--
Skim milk powder	133.33
Part B	--
Peptic digest of animal tissue	3.33
Sodium chloride	1.67
Yeast extract	1.00
Cetrимide	0.40
Agar	20.00
Final pH : 7.3 ± 0.2 at 25°C	
Formula may be adjusted and/or supplemented as required to meet performance specifications	





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Precautions

1. For Laboratory Use only.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.
3. Use the prepared medium as fresh as possible.

Directions

1. Suspend 26.4 grams of Part B in 250 ml distilled water.
2. Heat to boiling to dissolve the medium completely.
3. Autoclave at 121°C, 15 psi pressure, for 20 minutes / validated cycle.
4. Suspend 133.33 grams of Part A in 750 ml of distilled water and sterilize by autoclaving at 121°C, 15 psi pressure, for 5 minutes.
5. After autoclaving mix Part A and B and pour into sterile Petri plates.

Quality Control Specifications

Dehydrated Appearance	Part A : White to cream homogeneous free flowing powder Part B : Cream to yellow homogeneous free flowing powder
Prepared Medium	Light amber coloured opalescent gel forms in Petri plates
Reaction of 2.64% w/v aqueous solution of Part B	pH : 7.3 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 2.0% Agar gel

Expected Cultural Response: Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours.

Sr. No.	Organisms	Results to be achieved		
		Inoculum (CFU)	Growth	Pigment
1.	<i>Escherichia coli</i> ATCC 25922	>=10 ³	inhibited	--
2.	<i>Pseudomonas aeruginosa</i> ATCC 27853	50 -100	good-luxuriant	blue green
3.	<i>Stenotrophomonas maltophilia</i> ATCC 13637	>=10 ³	inhibited	--

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure

1. *P. aeruginosa* hydrolyzes casein and produces a yellowish to green diffusible pigment on Milk Agar. For isolation, filter 200ml or less water of the swimming pool through sterile membrane filters.
2. Place each membrane filter on M-PA Agar (DM608). Incubate the plates at 41.5 ± 0.5°C for 72 hours.
3. Typical *P. aeruginosa* colonies are 0.8-2.2 mm in diameter, flat in appearance with brownish to greenish centers.
4. For confirmation, using Milk Agar w/ Cetrimide, make a single streak from an isolated colony on a Milk Agar w/ Cetrimide plate and incubate at 35-37°C for 24 hours.
5. After incubation *P. aeruginosa* forms pigmented colonies.
6. Refer to appropriate references for standard test procedures.





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Results

P. aeruginosa forms yellowish green colonies on this medium. Cetrimide acts as a quaternary ammonium, cationic detergent that causes release of nitrogen and phosphorus from bacterial cells other than *P. aeruginosa*. Refer to appropriate references and test procedures for interpretation of results.

Storage

Store the sealed bottle containing the dehydrated medium at 2 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light.

Expiration

Refer to the expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedure

1. For identification, organisms must be in pure culture. Morphological, biochemical and/or serological tests should be performed for final identification.
2. Consult appropriate texts for detailed information and recommended procedures.

Packaging

Product Name : Milk Agar w/ Cetrimide (Twin Pack)

Product Code : DM349

Available Pack sizes : 100gm / 500gm

References

1. Brown M. R. W. and Scott F. J. H., 1970, J. Clin. Pathol., 23:172.
2. International Organization for Standardization (ISO), Draft ISO/DIS 8360-1:1988.

Further Information

For further information please contact your local MICROMASTER Representative.



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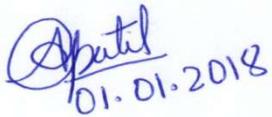
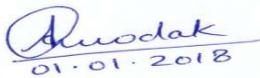
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