

## Technical Information

### PM Indicator Agar (Penicillin in Milk Indicator Agar)

#### Product Code: DM 1849

**Application:** PM Indicator Agar (Penicillin in Milk Indicator Agar) is recommended for rapid detection of trace amounts of Penicillin in milk using *Bacillus stearothermophilus* as per AOAC.

#### Composition\*\*

Ingredients	Gms / Litre
Peptic digest of animal tissue	5.000
Casein enzymic hydrolysate	1.700
Papaic digest of soyabean meal	0.3 00
Beef extract	3.000
Dextrose	5.250
Sodium chloride	0.500
Dipotassium phosphate	0.250
Polysorbate 80	1.000
Bromo cresol purple	0.060
Agar	15.000
Final pH ( at 25°C)	7.8±0.2

\*\*Formula adjusted, standardized to suit performance parameters

#### Principle & Interpretation

Through PM Indicator Agar is used for rapid detection of trace amounts of penicillin in milk yet AOAC has recommended *Bacillus stearothermophilus* qualitative discs <sup>(1)</sup>. PM Indicator Agar is designed as per the formula published by the 18th Annual Meeting of the National Mastitis Council <sup>(2)</sup>. This method is a modification of the procedure approved by the International Dairy Federation for the qualitative detection of penicillin in milk <sup>(3)</sup>. Originally a medium called Reductase Medium was suggested by Reid and Brewer for detecting penicillin in milk by using *Bacillus subtilis* <sup>(4)</sup>. The present medium is better, faster and more reliable than that of Reid and Brewer (as it support and demonstrate the growth and acid formation by *B. stearothermophilus*, which is sensitive to penicillin and beta-lactam residues.) To demonstrate the presence of traces of penicillin in milk; the qualitative disc method is found to be more suitable which is also recommended by AOAC.

Inoculate the medium with 1 ml of uniformly dispersed *B. stearothermophilus* suspension prepared as per AOAC <sup>(1)</sup>. Prepare dilutions of standard penicillin G (positive control) to give concentrations in the range of 0.005 to 0.1 units/ ml. Use a suspension of i



Dehydrated Culture Media  
Bases / Media Supplements

Clear zone >14mm	Clear zone 17-20mm	No zone	Positive test indicating that an inhibitor is present in test sample. Perform CONFIRMING ASSAY to determine if inhibitor is a beta-lactam residue.
Any reaction	Any reaction	Any zone	Result indicates error in the test system.
Any reaction	No zone	Any reaction	Determine source of error.
For Confirming Assay: Inactive milk sample by heating at 82°C for 2 minutes. Cool promptly to room temperature. Invert the plate and incubate as in screening assay.			
Interpretation of Confirming Assay			
Inactivated Test Sample Disc	Positive Control Disc	Penicillinase / Inactivated Milk Sample Disc	Interpretation
Clear zone >14 mm	Clear zone 17-20 mm	No zone	Positive test indicating the presence of beta - lactam residues.
Clear zone >14 mm	Clear zone 17-20 mm	Clear zone, same size as test Sample	Positive test indicating the presence of inhibitor(s) other than beta – lactam residues.
Clear zone >14 mm	Clear zone 17-20 mm	Clear zone substantially smaller than 14 mm	Positive test indicating presence of beta – lactam residues as well as other inhibitors.

## Methodology

Suspend 3.2 grams of powder media in 100 ml distilled water. Shake well and heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 50°C. Mix well and dispense as desired.

## Quality Control

### Physical Appearance

Beige to bluish grey homogeneous free flowing powder

### Gelling

Firm, comparable with 1.5% Agar gel

### Colour and Clarity of prepared medium

purple coloured clear to slightly opalescent gel forms in Petri plates

### Reaction

Reaction of 3.2% w/v aqueous solution at 25°C. pH : 7.8±0.2

pH Range 7.40-7.80

### Cultural Response/ characteristics

DM 1849: Cultural response observed after an incubation in the following parameters.

Organism	Growth	Incubation temperature	Time
Bacillus stearothermophilus ATCC 7953	good	55±2°C	3-4 hours
Bacillus stearothermophilus ATCC 7953	inhibited	64±2°C	3-4 hours



## Storage and Shelf Life

**Dried Media:** Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.

**Prepared Media:** 2-8° in sealable plastic bags for 2-5 days.

## Further Reading

1. Williams, (Ed.), 2005, Official Methods of Analysis of the Association of Official Analytical Chemists, 19th Ed., AOAC, Washington, D.C
2. Publ. of the 18th Annual Meeting of Natl. Mastitis Council, Inc.
3. International Dairy Federation, 1970, International Dairy Federation, Brussels, Belgium.
4. Reid R. D. and Brewer J. H., 1946, J. Bacteriol., 52: 251.

## Disclaimer :

- User must ensure suitability of the product(s) in their application prior to use.
- The product conform solely to the technical information provided in this booklet and to the best of knowledge research and development work carried at **CDH** is true and accurate
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