# Lactose Gelatin Medium

**M628**

## Intended Use

Recommended for detection of *Clostridium* species from food samples.

## Composition**

| Ingredients | Gms / Litre |
|---|---|
| Lactose | 10.000 |
| Disodium hydrogen phosphate | 5.000 |
| Gelatin | 120.000 |
| Phenol red | 0.050 |
| Final pH ( at 25°C) | 7.5±0.2 |

**Formula adjusted, standardized to suit performance parameters

## Directions

Suspend 135.0 grams in 1000 ml warm purified / distilled water. Heat if necessary to dissolve the medium completely and dispense 10 ml amounts in 15x150 mm screw capped tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Just before use, heat to boiling to remove dissolved oxygen and cool rapidly to incubation temperature.

## Principle And Interpretation

Members of the genus *Clostridium* are distributed widely in nature and are found in soil as well as in fresh water and marine water sediments throughout the world (5). Clostridial species are one of the major causes of food poisoning / gastro-intestinal illnesses. They are gram-positive, spore-forming rods that occur naturally in soil (1). Among the family are: *Clostridium botulinum*, which produces one of the most potent toxins in existence; *Clostridium tetani*, causative agent of tetanus; and *Clostridium perfringens*, commonly found in wound infections and diarrhoea cases. The use of toxins to damage the host is a method deployed by many bacterial pathogens including *Clostridium*.

Lactose Gelatin Medium is prepared as per APHA (6) for detecting *Clostridium* species from food samples. The medium contains lactose which is fermented by the *Clostridium* species, mainly by *Clostridium perfringens* yielding acid and gas. Phenol red is the pH indicator which turns yellow at acidic pH. Gelatin is usually liquefied by *Clostridium perfringens* within 24-48 hours (2). Disodium phosphate buffers the medium.

## Type of specimen

Food samples

## Specimen Collection and Handling

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (6). After use, contaminated materials must be sterilized by autoclaving before discarding.

## Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

## Limitations

1. Further isolation on selective media is required.
2. For complete identification, biochemical and serological tests must be carried out.

## Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

## Quality Control

**Appearance**

Light yellow to pink coloured homogeneous free flowing slightly coarse powder

**Colour and Clarity of prepared medium**

Red coloured clear to slightly opalescent gel forms in tubes.

**Reaction**
Reaction of 13.5% w/v aqueous solution at 25°C. pH : 7.5±0.2
**pH**
7.30-7.70
**Cultural Response**
Cultural characteristics observed under an anaerobic condition after an incubation at 35-37°C for 48-72 hours.

| Organism | Lactose Fermentation | Gelatin liquefaction |
|---|---|---|
| *Clostridium perfringens* ATCC 12924 | Positive reaction,yellow colour | Positive reaction |
| *Clostridium sporogenes* ATCC 11437 | Negative reaction,no colour change or red | Positive reaction |

## Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Product performance is best if used within stated expiry period.

## Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

## Reference

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3. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
4. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
5. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
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Revision : 03/2021