



MacConkey Agar, RS

M1702

Intended Use

For isolating and differentiating Gram negative enteric bacilli from specimens containing swarming strains of *Proteus* species.

Composition**

| Ingredients | g / L |
|---------------------|---------|
| Peptone | 17.000 |
| Proteose peptone | 3.000 |
| Lactose | 10.000 |
| Bile salts | 5.000 |
| Sodium chloride | 5.000 |
| Neutral red | 0.030 |
| Crystal violet | 0.001 |
| Agar | 13.500 |
| Final pH (at 25°C) | 7.1±0.2 |

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 53.53 grams in 1000 ml purified / distilled water. Heat to boiling with gentle swirling to dissolve the agar completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Avoid overheating. Cool to 45-50°C. Mix well and pour into sterile Petri plates. The surface of the medium should be dry when inoculated.

Principle And Interpretation

MacConkey agars are slightly selective and differential plating media mainly used for the detection and isolation of gram-negative organisms from clinical (1,2), dairy(3), food(4,5), water(6), and industrial sources (7). It is also recommended for the selection and recovery of the *Enterobacteriaceae* and related enteric gram-negative bacilli. These agar media is less selective since the concentration of bile salts, which inhibit gram-positive microorganisms, is low in comparison with other enteric plating media. Other than that this medium is also used for count of coli-aerogenes bacteria in cattle and sheep faeces (1), the count of coli-aerogenes and non-lactose fermenters in poultry carcasses (8), bacterial counts on irradiated canned minced chicken (9).

MacConkey Agar is the earliest selective and differential medium for cultivation of enteric microorganisms from a variety of clinical specimens (10,11). The original medium contains protein, bile salts, sodium chloride and two dyes. The selective action of this medium is attributed to crystal violet and bile salts, which are inhibitory to most species of gram-positive bacteria. Gram-negative bacteria usually grow well on the medium and are differentiated by their ability to ferment lactose. Lactose-fermenting strains grow as red or pink colonies and may be surrounded by a zone of acid precipitated bile. The red colour is due to production of acid from lactose, absorption of neutral red and a subsequent colour change of the dye when the pH of medium falls below 6.8. Lactose non-fermenting strains, such as *Shigella* and *Salmonella* are colourless, transparent and typically do not alter appearance of the medium.

Peptones are sources of nitrogen and other nutrients. Lactose is a fermentable carbohydrate, bile salts and crystal violet are selective agents that inhibit growth of gram-positive organisms. Neutral red is the pH indicator dye.

Type of specimen

Clinical samples - faeces, urine, pus; Food and dairy samples; Water samples, Pharmaceutical and industrial samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (1,2).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (3-5).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards(6).

For industrial samples, follow appropriate techniques for sample collection, processing as per guidelines (7).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

In Vitro diagnostic Use. For professional use only. 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 clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.
2. Each lot of the medium has been tested for the organisms specified on the COA. It is recommended to users to validate the medium for any specific microorganism other than mentioned in the COA based on the user's unique requirement.
3. Though the medium is recommended for selective isolation, further biochemical and serological tests must be carried out for complete identification.
4. The surface of the medium should be dry when inoculated.

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 homogeneous free flowing powder

Gelling

Firm comparable with 1.35% Agar gel.

Colour and Clarity of prepared medium

Orange red coloured, clear to slightly opalescent gel forms in Petri plates.

Reaction

Reaction of 5.35% w/v aqueous solution at 25°C. pH : 7.1±0.2

pH

6.90-7.30

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

| Organism | Inoculum (CFU) | Growth | Recovery | Colour of colony |
|---|------------------|--------------|----------|-----------------------------------|
| <i>Escherichia coli</i> ATCC 25922 (00013*) | 50-100 | luxuriant | ≥50% | pink to red with bile precipitate |
| # <i>Klebsiella aerogenes</i> ATCC 13048 (00175*) | 50-100 | luxuriant | ≥50% | pink to red |
| <i>Proteus vulgaris</i> ATCC 13315 | 50-100 | luxuriant | ≥50% | colourless |
| <i>Salmonella</i> Paratyphi A ATCC 9150 | 50-100 | luxuriant | ≥50% | colourless |
| <i>Shigella flexneri</i> ATCC 12022 (00126*) | 50-100 | fair to good | 30-40% | colourless |
| <i>Salmonella</i> Paratyphi B ATCC 8759 | 50-100 | luxuriant | ≥50% | colourless |
| <i>Salmonella</i> Enteritidis ATCC 13076 (00030*) | 50-100 | luxuriant | ≥50% | Colourless |
| <i>Salmonella Typhi</i> ATCC 6539 | 50-100 | luxuriant | ≥50% | colourless |
| <i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*) | ≥10 ⁴ | inhibited | 0% | |

Key : (*) Corresponding WDCM numbers.

(#) Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (1,2).

Reference

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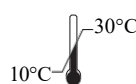
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