

MacConkey Broth, Granulated

GMH083

MacConkey Broth, granulated is used for the selective enrichment of *E.coli* from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP.

Composition**

Ingredients	Gms / Litre
Pancreatic digest of gelatin	20.000
Lactose monohydrate	10.000
Dehydrated ox-bile	5.000
Bromo cresol purple	0.010
pH after sterilization (at 25°C)	7.3±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 34.51 grams (the equivalent weight of dehydrated medium per litre) in 1000 ml purified/ distilled water. Heat if necessary to dissolve the medium completely. Dispense into test tubes with inverted Durham tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Principle And Interpretation

MacConkey Broth is a modification of MacConkey Medium (1). Childs and Allen (2) demonstrated the inhibitory effect of neutral red and therefore substituted it by the less inhibitory bromocresol purple dye. BCP is more sensitive in recording pH variation in the medium. This medium is prepared in accordance with the harmonized method of USP/BP/JP (4,5,6)

Pancreatic digest of gelatin provides essential growth nutrients. Lactose is the fermentable carbohydrate. Dehydrated ox-bile inhibits gram-positive organisms. Bromocresol purple is the pH indicator in the medium, which turns yellow under acidic condition. Lactose fermenting organisms turn the medium yellow due to the acidity produced on lactose fermentation. The colour change of the dye is observed when the pH of the medium falls below 6.8. Lactose non-fermenting organisms like *Salmonella* and *Shigella* do not alter the appearance of the medium.

Transfer homogenate in Soyabean Casein Digest Medium (GMH011/ MH011) containing 1 gm or 1 ml of the preparation to be examined to 100 ml MacConkey Broth Incubation is carried at 43°-45°C for 24-48 hours. For further isolation, subculture on MacConkey Agar (GMH081/ MH081). Growth of red generally non-mucoid colonies, sometimes surrounded by a reddish precipitation zone, indicates presence of coliforms.

Quality Control

Appearance

Cream to yellow with green tinge coloured granular medium

Colour and Clarity of prepared medium

Purple coloured clear to slightly opalescent solution in tubes

pH

7.10-7.50

Cultural Response

Growth Promotion is carried out in accordance with the harmonized method of USP/EP/BP/JP. For organisms not specified in pharmacopoeia, cultural response was observed after an incubation at 30-35°C for 18-48 hours.

Growth promoting properties

Clearly visible growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating ≤ 100 cfu (at 42-44°C for ≤ 24 hours).

Inhibitory properties

No growth of the test microorganism occurs for the specified temperature for not less than longest period of time specified inoculating ≥ 100 cfu (at 42-44°C for ≥ 48 hours).

Cultural Response

Cultural characteristics observed after an incubation at 30-35°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth	Acid	Gas	Incubation temperature	Incubation period
Growth promoting						
<i>Escherichia coli</i> ATCC 8739	50 -100	luxuriant	positive reaction, yellow colour	positive reaction	42 -44 °C	≤ 24 hrs
Inhibitory						
<i>Staphylococcus aureus</i> ATCC 6538	$\geq 10^3$	inhibited			42 -44 °C	≥ 48 hrs
Additional Microbiological testing						
<i>Escherichia coli</i> ATCC 25922	50 -100	luxuriant	positive reaction, yellow colour	positive reaction	30 -35 °C	18 -24 hrs
<i>Escherichia coli</i> NCTC 9002	50 -100	luxuriant	positive reaction, yellow colour	positive reaction	30 -35 °C	18 -24 hrs
<i>Enterobacter aerogenes</i> ATCC 13048	50 -100	luxuriant	positive reaction, yellow colour	positive reaction	30 -35 °C	18 -24 hrs
<i>Salmonella</i> Choleraesuis ATCC 12011	50 -100	fair-good	negative reaction	negative reaction	30 -35 °C	18 -24 hrs
<i>Staphylococcus aureus</i> ATCC 25923	$\geq 10^3$	inhibited			30 -35 °C	≥ 48 hrs

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label.

Reference

1. MacConkey A. T., 1900, The Lancet, ii: 20.
2. Childs E. and Allen, 1953, J. Hyg: Camb. 51:468-477.
3. European Pharmacopoeia, 2014, European Department for the Quality of Medicines of Council of Europe.
4. The United States Pharmacopoeia, 2014, The United States Pharmacopoeial Convention, Rockville, MD.
5. British Pharmacopoeia, 2014, The Stationery Office British Pharmacopoeia.
6. Japanese Pharmacopoeia, 2008, Published by Society of Japanese Pharmacopoeia, Tokyo, Japan.

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