



## PRODUCT SPECIFICATION SHEET

### MacConkey Broth (DM150I)

#### Intended Use

MacConkey Broth (DM150I) is recommended for the presumptive identification of coliforms from pharmaceutical products using the microbial limit testing in compliance with IP.

#### Product Summary and Explanation

MacConkey Broth is a modification of the original bile salt broth recommended by MacConkey that contained 0.5% sodium taurocholate and litmus as an indicator.<sup>(1)</sup> Further, MacConkey suggested variations of this formulation using neutral red indicator instead of litmus. Childs and Allen<sup>(2)</sup> 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. Oxgall in the medium replaces the original sodium taurocholate to inhibit growth of gram-positive organisms. This medium is recommended by Indian pharmacopoeia<sup>(3)</sup> for the presumptive identification of coliforms from water and other materials of sanitary importance. MacConkey Broth is used for cultivating gram-negative, lactose-fermenting bacilli and as a presumptive test for coliform organisms. It has been used to analyze food,<sup>(4)</sup> milk<sup>(5,6)</sup> and water samples<sup>(6-9)</sup> for coliforms.

#### Principles of the Procedure

MacConkey Broth contains gelatine peptone which provides essential growth nutrients for the growth microorganisms. Lactose is the fermentable source of carbohydrate. Dehydrated ox-bile inhibits gram-positive organisms. Bromocresol purple is the pH indicator in the medium, which turns the medium yellow under acidic condition produced by lactose fermenting organisms 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.

#### Formula / Liter

Ingredients	Gms / Liter
Gelatin Peptone	20.00
Lactose	10.00
Dehydrated ox-bile	5.00
Bromo cresol purple	0.01
Final pH: 7.3 ± 0.2 at 25°C	
Formula may be adjusted and/or supplemented as required to meet performance specifications	

#### Precautions

1. For Laboratory Use only.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

#### Directions

1. Suspend 35.01 grams of the medium in one liter of purified/ distilled water.
2. Heat if necessary to dissolve the medium completely.
3. Dispense into test tubes with inverted Durham tubes.
4. Autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.





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### Quality Control Specifications

Dehydrated Appearance	Cream to yellow homogeneous free flowing powder
Prepared Medium	Purple coloured clear to slightly opalescent solution in tubes
Reaction of % Solution	Not Applicable
Gel Strength	Not Applicable

### Cultural Response

Growth Promotion is carried out in accordance with the method of IP. 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  $\leq 100$  cfu (at 42-44°C for  $\leq 24$  hours).

### Inhibitory properties

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

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

Sr. No.	Organisms	Results to be achieved				
		Inoculum (CFU)	Growth	Acid	Gas	Incubation Temperature /Period
	<b>Growth promoting</b>					
1.	<i>Escherichia coli</i> ATCC 8739	50 -100	good-luxuriant	positive reaction, yellow colour	positive reaction	42 -44 °C $\leq 24$ hrs
	<b>Inhibitory</b>					
2.	<i>Staphylococcus aureus</i> ATCC 6538	$\geq 10^3$	inhibited	--	--	42 -44 °C $\geq 48$ hrs
	<b>Additional Microbiological Testing</b>					
3.	<i>Escherichia coli</i> ATCC 25922	50 -100	good-luxuriant	positive reaction, yellow colour	positive reaction	30 -35 °C 18 -24 hrs
4.	<i>Escherichia coli</i> NCTC 9002	50 -100	good-luxuriant	positive reaction, yellow colour	positive reaction	30 -35 °C 18 -24 hrs
5.	<i>Enterobacter aerogenes</i> ATCC 13048	50 -100	good-luxuriant	positive reaction, yellow colour	positive reaction	30 -35 °C 18 -24 hrs
6.	<i>Salmonella Choleraesuis</i> ATCC 12011	50 -100	fair-good	negative reaction	negative reaction	30 -35 °C 18 -24 hrs
7.	<i>Staphylococcus aureus</i> ATCC 25923	$\geq 10^3$	inhibited	--	--	30 -35 °C $\geq 48$ hrs

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





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### Test Procedure

1. Transfer homogenate in Soyabean Casein Digest Medium (DM277I) containing 1 gm or 1 ml of the preparation to be examined to 100 ml MacConkey Broth.
2. Incubate is carried at 43-45°C for 24-48 hours.
3. For further isolation, subculture on MacConkey Agar (DM143I).
4. Refer to appropriate references for standard test procedures.

### Results

1. Growth of red generally non-mucoid colonies, sometimes surrounded by a reddish precipitation zone, indicates presence of coliforms.
2. Refer to appropriate references and test procedures for interpretation of results.

### Storage

Store the sealed bottle containing the dehydrated medium at 10 - 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 :** MacConkey Broth

**Product Code :** DM150I

**Available Pack sizes :** 100gm/ 500gm

### References

1. MacConkey A. T., 1900, The Lancet, ii: 20.
2. Childs E. and Allen, 1953, J. Hyg: Camb. 51:468-477.
3. Indian Pharmacopoeia, 2014 Ministry of Health and Family Welfare, Govt. of India.
4. Qadri, Buckle and Edwards. 1974. J. Appl. Bact. 37:7-14.
5. Adeleke, Adeniyi and Akinrinmisi. 2000. Afr. J. Biomed. Res. 3:89-92.
6. Hsu and Tsen. 2001. Int. J. Food Microbiol. 64:1-11.
7. World Health Organization. 4 Sept 2008. European standards for drinking water, 2nd ed., online. [www.who.int/water\\_sanitation\\_health/dwq/eurostand2/en/index.html](http://www.who.int/water_sanitation_health/dwq/eurostand2/en/index.html).
8. Alivisatos and Papadakis. 1975. J. Appl. Bact. 39:287-293.
9. International Organization for Standardization. 1990. Water quality - Detection and enumeration of coliform organisms, thermotolerant coliform organisms and presumptive *Escherichia coli* - Part 2: Multiple tube (most probable number) method. ISO 9308-2, First ed., 1990-10-01. International Organization for Standardization, Geneva, Switzerland.

### Further Information

For further information please contact your local MICROMASTER Representative.





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