

For identification and differentiation of *E. coli* and Total coliforms

## HiCrome™ M-Lauryl Sulphate Agar

Recommended for the differentiation and enumeration of *Escherichia coli* and other coliforms by a single membrane filtration technique

M1569

### Composition \*\*

Ingredients	Grams/Litre
Peptone	40.00
Yeast extract	6.00
Lactose	30.00
Phenol red	0.20
Sodium lauryl sulphate (SLS)	1.00
Sodium pyruvate	0.50
Chromogen	0.20
Agar	10.00

Final pH (at 25°C) 7.4 ± 0.2

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

### Directions

Suspend 87.9 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45- 50°C. Mix well and pour into sterile Petri plates.

### Principle and Interpretation

HiCrome™ M-Lauryl Sulphate Agar is a modification of the Lauryl Tryptose Broth, formulated by Mallman and Darby, (1). This chromogenic medium is recommended for the presumptive identification and differentiation of *Escherichia coli* and other coliforms by a single membrane filtration technique (2, 3). The incorporation of chromogen X-glucuronide and the dye phenol red favours the differentiation of *E. coli* and other coliforms on the basis of colour.

Peptone provide nitrogenous and carbonaceous compounds, long chain amino acids and other essential growth nutrients to the organisms. Yeast extract serves as a source of vitamins especially group B vitamins. Lactose acts as a source of fermentable sugar while sodium lauryl sulphate inhibits gram positive organisms. The enzyme  $\beta$ -D-glucuronidase produced by *E. coli*, cleaves X-glucuronide, imparting a green colour to the colonies. Lactose fermentation is detected by phenol red indicator. Other lactose fermentors not possessing  $\beta$ -D-Glucuronidase enzyme will show yellow colonies, whereas lactose non-fermentors will exhibit pink coloured colonies.

### Type of specimen

Water samples

### Specimen Collection and Handling

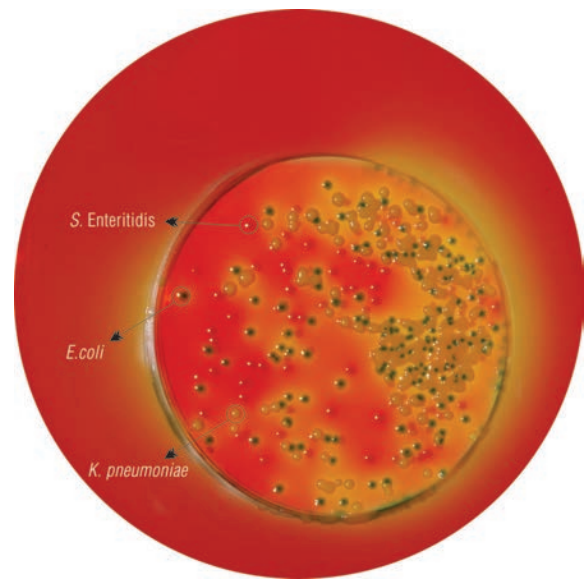
For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (4). After use, contaminated materials must be sterilized by autoclaving before discarding.

### 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.  $\beta$ -glucuronidase is present in 97% of *E. coli* strains, however few *E. coli* may be negative.
2. Some species may show poor growth due to nutritional variations.



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### Performance and Evaluation

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

### Quality Control

- Appearance of Powder** : Light yellow to pink coloured, homogeneous, free flowing powder.
- Gelling** : Firm, comparable with 1.0% Agar gel.
- Colour and Clarity of prepared medium** : Red coloured, clear to slightly opalescent gel forms in Petri plates.
- Reaction** : Reaction of 8.8% w/v aqueous solution at 25°C. pH:7.4 ± 0.2 .
- Cultural Response** : Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organisms (ATCC)	Inoculum (CFU)	Growth	Colour of colony
<i>Escherichia coli</i> (25922) (00013*)	50-100	luxuriant	green
<i>Klebsiella pneumoniae</i> (13883) (00097*)	50-100	good	yellow, mucoid
<i>Staphylococcus aureus</i> subsp. aureus (25923) (00034*)	≥10 <sup>3</sup>	inhibited	-
<i>Salmonella</i> Enteritidis (13076) (00030*)	50-100	good	pink

Key : \* : corresponding WDCM Numbers

### Storage and Shelf-life

Store between 2-8°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. Use before expiry date on the label.

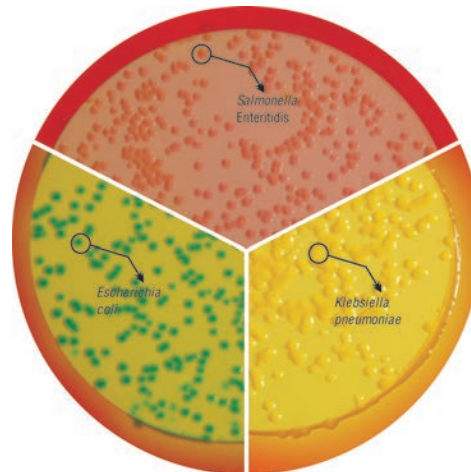
Product performance is best if used within stated expiry period.

### 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 (5, 6).

### References

1. Mallman and Darby, 1941, Am. J. Public Health, 31:127.
2. Methods for Examination of Waters and Associated Materials, Environment Agency, 1998, Standing Committee of Analysts.
3. Sartory D.P. and Howard L, 1992, Lett Appl. Microbiol. 15:273-276.
4. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. 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



M1569 – HiCrome™ M-Lauryl Sulphate Agar

Ready Prepared Media			
Code	Product Name	Usage	Packing
<b>Category : 55 mm Scored Polystyrene Plates</b>			
SP1569	HiCrome™ M-Lauryl Sulphate Agar Plate	for differentiation and enumeration of <i>Escherichia coli</i> and other coliforms by single membrane filtration	100 plts