

Harlequin® E. coli/Coliform Agar (NCM1002)

Intended Use

E. coli / Coliform Agar is for the simultaneous enumeration of *Escherichia coli* and coliforms in food and environmental samples and is not intended for use in the diagnosis of disease or other conditions in humans.

Description

This dual chromogenic substrate medium makes the different colony types easy to distinguish allowing rapid counting of both *E. coli* and coliforms on a single agar.

Based upon the formulation of Tryptone Bile Agar, the medium has been modified by the addition of two chromogenic substrates, one to detect the β -glucuronidase enzyme (X-glucuronide) and another to detect the β -galactosidase enzyme (magenta- β -gal). Typical *E. coli* strains possess both enzymes but only cleave the X-glucuronide substrate, thereby producing blue-green colonies. Typical coliforms, however, possess only the β -galactosidase enzyme and produce rose-pink colonies.

The colony types are easily distinguishable, even in the presence of other organisms, or when large numbers are observed, making simultaneous enumeration of *E. coli* and coliforms a quick and simple procedure.

Typical Formulation

Tryptone	20.0 g/L
Bile Salt No.3	1.5 g/L
X-glucuronide	0.075 g/L
Magenta- β -Galactoside	0.1 g/L
Agar	15.0 g/L

Final pH: 7.2 \pm 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precaution

Refer to SDS

Preparation

1. Suspend 36.6 grams of the medium in one liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave for 15 minutes at 121°C.
4. Cool to 45-50°C.

Test Procedure

- Inoculate 0.5 ml of a 1:10 dilution of the sample and spread over the entire surface of the plate. Further dilution may be necessary if large numbers of *E. coli* and/or coliforms are present, to ensure colonies can be easily counted.
- Incubate at 37°C for 18-24 hours.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, free-flowing and beige.

Prepared Appearance: Prepared medium is clear and yellow.



Technical Specification Sheet



Minimum QC:

Escherichia coli ATCC 25922

Enterobacter aerogenes ATCC 13048

Staphylococcus aureus ATCC 25923 (inhibited)

Results

Count all dark blue to violet colonies as presumptive *E. coli*, and calculate the cfu/g. Count all rose-pink colonies as presumptive coliforms, and calculate cfu/g.

Organism	Colony Size (mm)	Color
<i>Escherichia coli</i>	0.1 – 2.0	Dark blue to violet
<i>Enterobacter aerogenes</i>	1.5 – 2.5	Rose-pink
<i>Pseudomonas aeruginosa</i>	0.5 – 1.0	Beige
<i>Enterococcus faecalis</i>	No growth	
<i>Staphylococcus aureus</i>	No growth	

Expiration

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing or appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedures

Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

Storage

Store dehydrated culture media at 2-30°C away from direct sunlight. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

References

1. Baylis, C.L., Patrick, M. (1999). *Comparison of a range of Chromogenic media for enumeration of total Coliforms and Escherichia coli in foods*. Leatherhead International Technical Notes No.135: 99.



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