



HiCrome ECC Agar

M1293

HiCrome ECC Agar is a differential medium recommended for presumptive identification of *Escherichia coli* and other coliforms in food and environmental samples.

Composition**

Ingredients	Gms / Litre
Peptone, special	5.000
Yeast extract	3.000
Lactose	2.500
Disodium hydrogen phosphate	3.500
Monopotassium dihydrogen phosphate	1.500
Sodium chloride	5.000
Chromogenic mixture	20.300
Neutral red	0.030
Agar	15.000
Final pH (at 25°C)	6.8±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 55.83 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 50°C. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Escherichia coli, a member of the family *Enterobacteriaceae* is a part of normal flora of the intestinal tract of humans and a variety of animals. Although most of *E. coli* does not cause gastrointestinal illnesses, certain groups of *E. coli* can cause life-threatening diarrhoea and sever sequelae or disability (1). HiCrome ECC Agar is a differential medium recommended for the presumptive identification of *E. coli* and other coliforms in food and environmental samples (2). The medium contains two chromogens. One of the chromogen is cleaved by the enzyme glucuronidase produced by *E. coli* to give blue to purple coloured colonies whereas the other chromogen is cleaved by the enzyme galactosidase, produced by majority of coliforms, resulting in the formation of rose-pink coloured colonies (3, 4).

Peptone special, yeast extract provide nitrogenous substances, vitamin B complex and other essential growth nutrients. Lactose is the fermentable carbohydrate, which aids in detecting lactose fermenters with neutral red as an indicator. Disodium hydrogen phosphate and potassium dihydrogen phosphate buffers the medium well. Sodium chloride maintains the osmotic equilibrium. Dry the surface of plate medium.

Dilute the food sample by 1: 5 or 1: 10 with 0.1% sterile Peptone Water (M028) and homogenize in a blender or a stomacher. Spread 0.5 ml or 1.0 ml of the homogenate over the agar surface with a sterile glass spreader and incubate the plates at 37°C for 18-24 hours. Count the blue/purple colonies and multiply with the dilution factor. The number of *E. coli* is reported per gram of food. The medium should be used only for in-vitro diagnostic purpose. Wear mask while handling the dehydrated product and avoid contact with eyes.

Quality Control

Appearance

Light yellow to pink homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Reddish pink coloured, opaque gel forms in Petri plates

Reaction

Reaction of 5.58% w/v aqueous solution at 25°C. pH : 6.8±0.2

pH

6.60-7.00

Cultural Response

M1293: 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	50-100	luxuriant	≥70%	blue/purple
<i>Klebsiella pneumoniae</i> ATCC 13883	50-100	luxuriant	≥70%	rose/pink
<i>Pseudomonas aeruginosa</i> ATCC 27853	50-100	good-luxuriant	≥70%	straw
<i>Salmonella Enteritidis</i> ATCC 13076	50-100	luxuriant	≥70%	pink

Storage and Shelf Life

Store dehydrated powder and prepared medium at 2-8°C. Use before expiry period on the label.

Reference

1. Doyle M. P., (Ed.), 1989, Foodborne Bacterial Pathogens, Marcel Dekker, New York
2. Frampton E.W., Restaino L. and Blaszkowski N., 1988, J. Food Prot., 51:402.
3. Kilian M. and Bülow P., 1976, Acta. Pathol. Microbiol. Scand., Sect. B, 84:245.
4. Kilian M. and Bülow P., 1979, Acta. Pathol. Microbiol. Scand., Sect. B, 87:271.

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