# HiCrome ECC Selective HiVeg Agar Base

**MV1294**

HiCrome ECC Selective HiVeg Agar Base is recommended for detection of *Escherichia coli* and coliforms in water and food samples.

## Composition**

| Ingredients | Gms / Litre |
|---|---|
| HiVeg special peptone | 6.000 |
| HiVeg hydrolysate | 3.300 |
| Sodium dihydrogen phosphate | 0.600 |
| Disodium hydrogen phosphate | 1.000 |
| Sodium chloride | 2.000 |
| Sodium pyruvate | 1.000 |
| L-Tryptophan | 1.000 |
| Sorbitol | 1.000 |
| Tergitol 7 | 0.150 |
| Chromogenic mixture | 0.430 |
| Agar | 10.000 |
| Final pH ( at 25°C) | 6.8±0.2 |

**Formula adjusted, standardized to suit performance parameters

## Directions

Suspend 26.48 grams in 1000 ml distilled water. Heat in a boiling water bath or in a free flowing steam, with stirring to dissolve the medium completely (approximately 35 minutes). DO NOT AUTOCLAVE OR OVERHEAT. If desired, selective medium can be prepared by aseptically adding the rehydrated contents of 1 vial of HiCrome ECC Selective Supplement (FD190) to previously cooled sterile medium. Mix well and pour into sterile Petri plates. Medium may show haziness, but it does not affect the performance of the medium.

## Principle And Interpretation

HiCrome ECC Selective HiVeg Agar Base is a slight modification of HiCrome ECC Selective Agar and is prepared by replacing animal based peptones with vegetable peptones. HiCrome ECC Selective Agar is a selective medium recommended for the simultaneous detection of *Escherichia coli* and total coliforms in water and food samples (1, 2). The chromogenic mixture contains two chromogenic substrates. The enzyme beta-D-galactosidase produced by coliforms cleaves one of the chromogen to form salmon to red coloured colonies (3). The enzyme beta-D-glucuronidase produced by *E. coli* , cleaves X-glucuronide, the other chromogen (4). Colonies of *E. coli* give dark blue to violet coloured colonies due to cleavage of both the chromogens. Addition of L-Tryptophan improves the indole reaction, thereby increasing the detection reliability. HiVeg special Peptone, sodium pyruvate and sorbitol provide nitrogenous substances, fermentable carbohydrate and other essential growth nutrients for the organisms. Phosphates buffer the medium. The media formulation helps even sublethally injured coliforms to recover and grow rapidly. Tergitol inhibits gram-positive as well as some gram-negative bacteria other than coliforms (3). Addition of HiCrome ECC Selective Supplement (FD190) helps to inhibit the accompanying heterogenous microflora.

The medium is inoculated either by pour plate technique or by spreading the sample on the surface of plated medium. Membrane filter technique can also be used. To confirm *E. coli* , add a drop of Kovacs reagent on the dark blue to violet colony. Formation of cherry red colour indicates a positive reaction.

## Quality Control

**Appearance**

Light yellow to pink homogeneous free flowing powder

**Gelling**

Firm, comparable with 1.0% Agar gel.

**Colour and Clarity of prepared medium**
Light pink coloured, clear to slightly opalescent gel forms in Petri plates

**Reaction**
Reaction of 2.65% w/v aqueous solution at 25°C. pH : 6.8±0.2

**pH**
6.60-7.00

**Cultural Response**
Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

**Cultural Response**

| Organism | Inoculum (CFU) | Growth | Recovery | Colour of Colony | Indole production |
|---|---|---|---|---|---|
| **Cultural Response** | | | | | |
| *Citrobacter freundii ATCC 8090* | 50-100 | luxuriant | >=50% | salmon to red (big) | negative reaction |
| *Enterobacter aerogenes ATCC 13048* | 50-100 | luxuriant | >=50% | salmon to red | negative reaction |
| *Escherichia coli ATCC 25922* | 50-100 | good-luxuriant | >=50% | dark blue to violet | positive reaction |
| *Enterococcus faecalis ATCC 29212* | >=$10^3$ | inhibited | 0% | | |
| *Salmonella Enteritidis ATCC 13076* | 50-100 | good | 40-50% | colourless | negative reaction |
| *Shigella flexneri ATCC 29508* | 50-100 | good | 40-50% | light blue to turquoise | negative reaction |
| *Escherichia coli O157:H7 NCTC 12900* | 50-100 | luxuriant | >=50% | salmon to red | positive reaction |

## Storage and Shelf Life
Store dehydrated and prepared medium at 2-8°C in tightly closed container. Use before expiry period on the label.

## Reference
1.Frampton E.W., Restaino L. and Blaszko N., 1988, J.Food Prof., 51:402.
2.Kilian M. and Bülow P., 1976, Acta. Pathol. Microbiol. Scand Sect. B, 84:245.
3.LeMinor L. and Hamida F., 1962, Ann. Inst. Pasteur > 102:267.
4.Manafi M. and Kneifel W., 1989, Zentralbl. Hyg., 189:225.

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