



Reference : 01-618

Scharlau Microbiology - Technical data sheet

Product :

**Microinstant® CHROMOGENIC COLINSTANT  
AGAR**

### Specification

Solid selective and differential medium for the detection of total coliforms and *E. coli* in water and food samples.

### Formula \* in g/L

Tryptone.....	10.00
Yeast extract.....	3.00
Meat extract.....	5.00
Bile salts No. 3.....	1.50
Di-sodium phosphate.....	2.70
Sodium phosphate.....	2.20
Chromogenic mixture.....	0.40
Agar.....	13.00

Final pH 6,8 ±0,2 at 25 °C

\* Adjusted and /or supplemented as required to meet performance criteria

### Directions

Suspend 37,8 g of powder in 1 L of distilled water. Bring to the boil and distribute into suitable containers. Sterilize in the autoclave at 121°C for 15 minutes. If the medium is used on the same day of preparation sterilization can be omitted, but the boiling must be maintained for 2-3 minutes.

Note: This is a low selectivity medium. If high numbers of accompanying microbiota, especially *Pseudomonas* and *Aeromonas* are expected, the selectivity of the medium can be improved by the addition of the contents of one vial of Coliform CV Selective Supplement (Art. No. 06-140LYO1) to 500 mL of the medium cooled at 45-50°C.

### Description

The selectivity of Colinstant Chromogenic Agar is due to the surfactant action of bile salts No.3 that inhibits the growth of almost all Gram positive bacteria. Other inhibitory agents must be added if a greater selectivity is desired.

The chromogenic mixture is mainly composed of two substances: 6-chloro-3-indolyl-β-D-galacto-pyranoside and 5-bromo-4-chloro-3-indolyl-β-D-glucuronide. The first is cleaved by the characteristic enzyme of coliforms, β-D-galactosidase and gives a salmon to red colour to the coliform colonies. The second chromogenic substance is cleaved by the β-D-glucuronidase enzyme characteristic of *E. coli* and turns the colonies a blue colour.

*E. coli* has both enzymes and cleaves both chromogenic substances which produces a dark blue to violet colonies. Total coliforms are the sum of *E. coli* colonies plus the salmon-red colonies. Other Gram negative bacteria produce colourless colonies except for some that possess glucuronidase activity (but not galactosidase) and produces light blue to turquoise colonies.

To confirm the *E. coli* colonies it is recommended to verify by the production of indol: coat the blue-violet colonies with a drop of Kovacs' Reagent (Art. No. RE0007). If the reagent turns a cherry-red colour in a few seconds this is a positive production of indol which confirms the presence of *E. coli*.

If the Colinstant Chromogenic Agar is used with the membrane filter method, the colour and growth of the colonies may be effected. It is advisable to perform validation of the membrane filter used.

### Necessary supplements

Coliform CV Selective Supplement (Art. No. 06-140LYO1)

Vial Contents:

Necessary amount for 500 mL of complete medium.

Cefsulodin..... 2,50 mg

Vancomycin..... 2,50 mg

Distilled water (Solvent)



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### Quality control

**Incubation temperature:** 36°C ±2.0

**Incubation time:** 18-24 h

**Inoculum:** Practical range 100 ± 20 CFU. Min. 50 CFU (Productivity) / 10<sup>4</sup>-10<sup>6</sup> CFU (Selectivity) / ≥ 10<sup>3</sup> CFU (specificity) according to ISO 11133:2014/Amd 1:2018 . MF methods.

### Microorganism

*Escherichia coli* ATCC® 25922

*Escherichia coli* ATCC® 8739

*Salmonella enterica* ATCC® 13076

*Pseudomonas aeruginosa* ATCC® 10145

*C.freundii* ATCC® 43864

*Enterococcus faecalis* ATCC® 19433

### Growth

Productivity > 0.70

Productivity > 0.70

Productivity > 0.70

Good (Specificity)

Productivity > 0.70

Inhibited

### Remarks

Colonies: Blue-violet

Colonies: Blue-violet

Colonies: Colorless

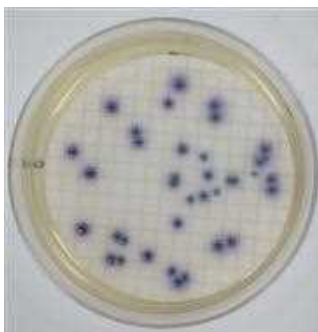
Colonies: Colorless

Colonies: Salmon to red

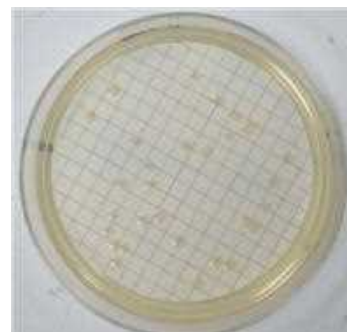
w. Selective Supplement



*Escherichia coli* ATCC 25922



*Escherichia coli* ATCC 8739



*Salmonella enteritidis* ATCC 13076

### References

- ADAMS, M., R.GRUBB, S.M. HAMER & A. CLIFFORD (1990) Colorimetric enumeration of *Escherichia coli* based on β-glucuronidase activity. *Appl. Environ. Microbiol.* 56:2021.
- KILIAN, M. & P. BÜLOW (1976) Rapid Diagnostic of Enterobacteriaceae. I. Detection of bacterial glycosidases. *Acta Pathol. Microbiol. Scand. Sect. B* 84:245:251.
- ISO 11133:2014/ Adm 1:2018. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.

### Storage

For laboratory use only. Keep tightly closed, away from bright light, in a cool dry place (+4 °C to 30 °C).