



ChromoCult[®] Coliform Agar ES

For detection of coliforms
in waste water and fresh foods.



ChromoCult[®]

Coliform Agar ES



Cat.No. 1.00850.0500 (500 g)

Selective agar for the simultaneous detection and colony count of total coliforms and E.coli in fresh foods as well as in waste water samples.

AOAC approval pending

Mode of action

The combination of carefully selected peptones and the buffering capacity of MOPS create ideal conditions for rapid growth of coliforms and support an optimal transformation of the chromogenic substrates. Bile salts and propionate largely inhibit growth of accompanying Gram-positive and Gram-negative flora.

The simultaneous detection of total coliforms and E.coli is achieved using the combination of two specific chromogenic substrates. The substrate Salmon[™]-β-D-GAL is split by β-D-galactosidase, characteristic for coliforms, resulting in a salmon to red colouration of coliform colonies. Detection of the β-D-glucuronidase characteristic for E.coli is achieved via cleaving the substrate X-β-D-glucuronide, resulting in a blue colouration of positive colonies.

As E.coli splits both Salmon[™]-β-D-GAL as well as X-β-D-glucuronide, the colonies turn a dark-violet colour and can be easily differentiated from the other salmon-red coliform colonies.

Typical composition (g/Litre)

Peptones 5,0; potassium chloride 7,5; MOPS 10,0; bile salts 1,15; propionate 0,5; Agar-agar 10,0; 6-Chloro-3-indoxyl-beta-D-galactopyranoside 0,15; isopropyl-beta-D-thiogalactopyranoside 0,1; 5-bromo-4-chloro-3-indoxyl-beta-D-glucuronic acid 0,1.

Preparation

Suspend 34.5 g in 1000 ml of demin. water and heat to boiling with frequent agitation until completely dissolved (approximately 45 minutes).

Do not autoclave, do not overheat!

The medium is cooled to 45 - 50 °C in a water bath and poured into sterile plates.

Note: a precipitate may appear if held for over 2 hours at this temperature.

pH: 7.0 ± 0.2 at 25 °C.

The plates are clear and colourless. When stored at +4 °C ± 2 °C, the shelf life of poured plates is 2 weeks.

Sample preparation

It is recommended to use a 1:10 dilution of the sample in a buffered solution (e.g. Peptone Water buffered or Sodium chloride peptone broth buffered) to prevent an interference between the colouration of coliform and E.coli colonies and the sample (e.g. low pH).

Application

Inoculate the medium using the pour-plate method, surface spreading or membrane-filter technique. The type of membrane filter affects the performance of the medium (growth and colouration of colonies). Best results are obtained using membrane filters of cellulose-mixed-ester material, e.g. Pall GN 6 (OSSMER, 1999).

Incubation: 24 hours at 35–37 °C.

Results

E.coli: dark blue to violet colonies (Salmon[™]-β-D-GAL and X-β-D-glucuronide reaction). Some E.coli (3 - 4 %) are β-glucuronidase-negative and grow as salmon-red colonies e.g. E.coli O157 strains.

Total coliforms: salmon to red colonies (Salmon[™]-β-D-GAL reaction) and dark blue to violet colonies (E.coli).

Accompanying flora: colourless/turquoise colonies.



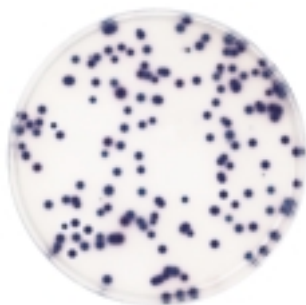


Colour makes the difference.

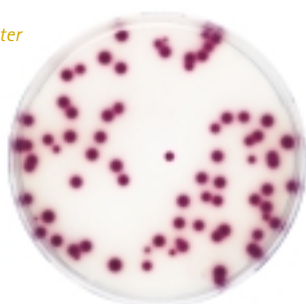
Quality control

Test strains	Inoculum (cfu/plate)	Growth	Colony colour
<i>Escherichia coli</i> ATCC 11775	30 – 300	+	dark blue to violet
<i>Citrobacter freundii</i> ATCC 8090	30 – 300	+	salmon-red
<i>Enterobacter cloacae</i> ATCC 13047	30 – 300	+	salmon-red
<i>Salmonella typhimurium</i> ATCC 14028	30 – 300	+	colourless
<i>Aeromonas hydrophila</i> ATCC 7966	1000 – 2000	-	
<i>Serratia liquefaciens</i> ATCC 27592	1000 – 2000	-	
<i>Staphylococcus aureus</i> ATCC 25923	1000 – 2000	-	
<i>Lactococcus lactis</i> ATCC 19435	1000 – 2000	-	
<i>Bacillus subtilis</i> ATCC 6633	1000 – 2000	-	

E.coli



Citrobacter



Literature

FRAMPTON, E.W.; RESTAINO, L. and BLASZKO, L.:

Evaluation of the β -glucuronidase substrate 5-bromo-4-chloro-3-indolyl- β -D-glucuronide (X-GLUC) in a 24 hour direct plating method for Escherichia coli.

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• *Espanola de Microbiologie, Granada, Spain* (1999)

KILIAN, M. and BÜLOW, P.:

Rapid diagnosis of Enterobacteriaceae. I. Detection of bacterial glycosidases.

• *Acta Pathol. Microbiol. Scand. Sect. B* 84:245–251 (1976)

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Avantages de la recherche de la β -galactosidase sur celle de la fermentation du lactose en milieu complexe dans le diagnostic bactériologique, en particulier des Enterobacteriaceae.

• *Ann. Inst. Pasteur (Paris)* 102:267–277 (1962)

MANAFI, M. and KNEIFEL, W. A.:

Combined chromogenic-fluorogenic medium for the simultaneous detection of total coliforms and E.coli in water.

• *Zentralbl. Hyg.* 189:225–234 (1989)

Additives

Merck Cat.No.	Product	Pack size
1.10582.0500	Sodium chloride peptone broth (buffered)	500 g
1.07228.0500	Peptone Water (buffered)	500 g



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Merck KGaA
64271 Darmstadt, Germany
Fax +49 (0) 61 51/72 33 80
E-mail: mibio@merck.de
Internet: microbiology.merck.de

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