

# Chromocult® Enterococci Agar

Selective culture medium for the isolation, differentiation and enumeration of Enterococci in water, foodstuffs and other materials.

## Mode of Action

The presence of Enterococci, especially *E. faecalis*, *E. faecium*, *E. durans* and *E. hirae*, serves as an indicator for faecal contamination.

Growth of Enterococci is stimulated by selected peptones, phosphates and addition of Tween® 80. Enterococci cleave the unique chromogenic substrates in the medium. This produces red colonies allowing an easy detection of Enterococci.

Sodium azide and ox bile inhibit most accompanying microbial flora. Non-Enterococci produce colourless, blue/violet or turquoise colonies. These colonies are easily distinguished from the red coloured colonies Enterococci produce.

## Typical Composition (g/litre)

Peptones 10.0; sodium chloride 5.0; sodium azide 0.2; di-potassium hydrogenphosphate 3.4; potassium di-hydrogen-phosphate 1.6; ox bile 0.5; Tween® 80 1.0; chromogenic-mixture 0.25; agar-agar 11.0

## Preparation

Suspend 33.0 g in 1 litre of demin. water by heating in a boiling water bath or in a flowing steam. Stir the contents to assist dissolution (approx. 45 minutes), let the medium cool to 45-50 °C and pour into plates.

■ Do not autoclave! Do not overheat!

pH. 7.0 ± 0.2 at 25 °C

The plates are clear and slightly yellow. If stored at +4 ± 2 °C and protected from light the plates are stable for 2 weeks.

## Experimental Procedure

Inoculate the medium by the pour-plate-method or by spreading the sample material on the surface of the plates. In addition the membrane-filter-technique can also be used.

The type of membrane filter affects the performance of the medium (growth and colouration of colonies). Best results were obtained using membrane filters of cellulose-mixed-ester material, e.g. Gelman GN-6 (OSSMER, 1999).

Incubation: 24 ± 4 hours at 35-37 °C.

If this will neither result in a colour change nor in visible growth continue the incubation up to 44 ± 4 hours.

## Evaluation

Enterococci:

Red colonies with a diameter of 0.5 to 2 mm

Non-Enterococci:

colourless (e.g. *Aerococcus viridans* ATCC 29503)

blue/violet (e.g. *Aerococcus viridans* ATCC 10400)

turquoise (e.g. *Streptococcus equi* ATCC 33398)

## Literature

DOTT, H. W., HAVEMEISTER, G., MÜLLER, H. E. and SACRÉ, C. 1982, Faecal streptococci as indicator organisms of drinking water. – *Zbl. Bakt. Hyg., I. Abt. Orig. A* 252; 154-165

OSSMER, R., SCHMIDT, W., MENDE, U. 1999, Chromocult® Coliform Agar – Influence of Membrane Filter Quality on Performance. – Poster presentation Congreso de la Sociedad, Espanola de Microbiologia, Granada, Spain

AMOROS, I. 1995, Evaluation of Chromocult® Enterococci Broth (with Agar) – Poster presentation Congress of Spanish Society of Microbiology, Madrid

LITSKY, W., MALLMANN, W. L. and Fifield, C. W. 1953, A new medium for the detection of enterococci in water. – *Amer. J. Pbl. Hlth.* 43; 873-879

MANAFI, M. and Windhager, K. 1997, Rapid identification of enterococci in water with a new chromogenic assay. – *Abstr. P-107*, pp. 453, Abstracts of the 97<sup>th</sup> Meeting of the American Society for Microbiology, Miami, USA

SNYDER, M. L. and LICHSTEIN, H. C. 1940, Sodium azide as an inhibiting substance for gram-negative bacteria. – *J. Infect. Dis.* 67, 113-115

## Ordering Information

Product	Merck Cat. No.	Pack size
Chromocult® Enterococci Agar	1.00950.0500	500 g

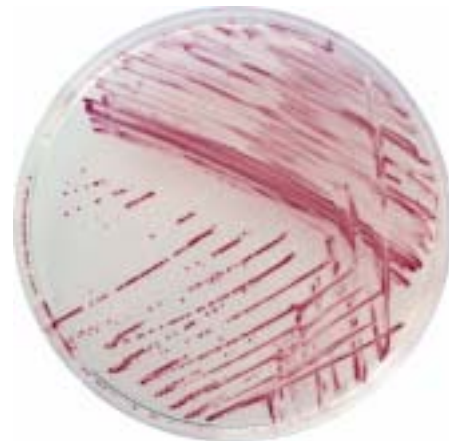
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## Quality control

Test strains	Inoculum (c.f.u./plate)	Growth	Colony colour
<i>Enterococcus faecalis</i> ATCC 19433	30 – 300	good	red
<i>Enterococcus faecium</i> ATCC 882	30 – 300	good	red
<i>Enterococcus durans</i> ATCC 6056	30 – 300	good	red
<i>Enterococcus hirae</i> ATCC 8043	30 – 300	good	red
<i>Aerococcus viridans</i> ATCC 10400	1000 - 2000	fair / none	blue / violet
<i>Bacillus cereus</i> ATCC 11778	1000 - 2000	-	-
<i>Escherichia coli</i> ATCC 11775	1000 - 2000	-	-
<i>Pseudomonas aeruginosa</i> ATCC 27853	1000 - 2000	-	-



*Aerococcus viridans*  
ATCC 10400



*Aerococcus faecalis*  
ATCC 19433