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 SACRÈ, C. 1982, Faecal streptococci as indicator organisms of drinking water. – Zbl. Bakt. Hyg., I. Abt. Orig. A 252; 154-165

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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 97th 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
Enterococcus faecalis ATCC 19433	30 – 300	good	red
Enterococcus faecium ATCC 882	30 – 300	good	red
Enterococcus durans ATCC 6056	30 – 300	good	red
Enterococcus hirae ATCC 8043	30 – 300	good	red
Aerococcus viridans ATCC 10400	1000 - 2000	fair / none	blue / violet
Bacillus cereus ATCC 11778	1000 - 2000	-	-
Escherichia coli ATCC 11775	1000 - 2000	-	-
Pseudomonas aeruginosa ATCC 27853	1000 - 2000	-	-

Aerococcus viridans
ATCC 10400

Aerococcus faecalis
ATCC 19433