

HiCrome™ VRE Agar Base / Modified

 $Recommended \ for \ identification \ of \ Vancomycin \ Resistant \ Enterococci \ from \ clinical \ specimens$

کار	M1830/ L
出	M1925

Composition **		
	M1830	M1925
Ingredients	Grams/Litre	Grams/Litre
Peptone special	25.00	20.00
Chromogenic mixture	0.45	3.60
Sodium chloride	5.00	5.00
Buffering agent	1.25	_
Salt mixture	4.25	_
Arabinose	_	10.00
Phenol red	_	0.10
Agar	15.00	15.00
Final pH (at 25°C)	6.5 ± 0.2	7.80 ± 0.2

^{**} Formula adjusted, standardized to suit performance parameters

Directions

Suspend 50.95 grams of (M1830) and 53.70 grams of (M1925) in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and aseptically add the rehydrated contents of two vials of HiCrome™ VRE Agar Supplement (FD277). Mix well and pour into sterile Petri plates.

Principle and Interpretation

Enterococci are the common habitants of the normal flora residing in the intestines of mammals (1). Vancomycin Resistant Enterococci are the group of Enterococci that have developed resistance towards many antibiotics particularly vancomycin. Enterococcal infections that result in human disease can be fatal, particularly those caused by strains of vancomycin-resistant enterococci (VRE) (2). Early detection of VRE is important to prevent the emergence of vancomycin resistance in *Enterococcus faecalis*.

VRE can be transmitted from person to person, especially in a hospital or chronic-care facility. Microscopic amounts of fecal material from an infected or colonized patient can contaminate the hospital environment and be a reason for the spread of infection. There are many traditional media for the detection of VRE which includes Vancomycin Resistant Enterococci Broth Base/ Agar or Bile Esculin Agar supplemented with vancomycin.

Peptone special in the medium supplies nitrogenous and carbonaceous compounds, long chain amino acids, vitamins and other necessary nutrients required for the growth of microorganisms. Sodium chloride maintains the osmotic balance. Buffering agents provides buffering to the medium. *Enterococcus faecalis* cleaves the chromogenic substrate in the medium to produce blue coloured colonies, which are clearly visible against the opaque background. The supplement added to the medium allows the selective isolation of Vancomycin Resistant Enterococci. This medium can be inoculated directly from screening swab, isolated colony prepared as a liquid suspension approximately equivalent to 0.5 McFarland turbidity.

Enterococcus faecium ferments arabinose and cleaves the substrate

thereby producing green colonies with yellow background. *Enterococcus faecalis* does not ferment arabinose thereby producing blue colonies due to cleavage of chromogenic substrate. The supplement added to the medium allows the selective isolation of Vancomycin Resistant Enterococci. This medium can be inoculated directly from screening swab, isolated colony prepared as a liquid suspension approximately equivalent to 0.5 McFarland turbidity.

Type of specimen

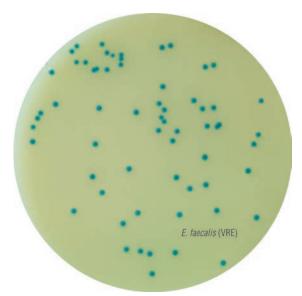
Clinical samples

Specimen Collection and Handling

For clinical samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (3, 4). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

In Vitro diagnostic use only. Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets



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Limitations

- Some Intermediate species may show poor growth due to nutritional variations and tolerance to vancomycin.
- Slight colour variation may be observed depending upon the utilization of the substrate by the organism.
- 3. Further confirmation has to be carried using sensitivity testing.
- 4. For M1830, interspecies differentiation between *Enterococcus faecalis* and *Enterococcus faecium* cannot be confirmed.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the recommended temperature.

Quality Control

Appearance of Powder: Cream to yellow (M1830) or Light yellow to pink (M1925) homogeneous free flowing

powder

Gelling Colour and Clarity

Colour and Clarity of prepared medium Reaction Firm, comparable with 1.5% Agar gel.
Off white coloured (M1830) or red coloured (M1925) opaque gel forms in Petri plates.

: Reaction of 5.1% w/v (M1830) or 5.37% w/v (M1925) aqueous solution at 25°C.

pH : 6.5 ± 0.2 (M1830) pH : 7.8 ± 02 (M1925)

Cultural Response

 Cultural characteristics observed with added HiCrome™ VRE Agar Supplement (FD277) after an incubation at 35-37°C for 24-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony (M1830)	Colour of colony (M1925)
Enterococcus faecalis (VRE) (51299) (00085*) (00152*)	50-100	luxuriant	≥50%	bluish green	blue
Enterococcus faecium (VRE) (700221)	50-100	luxuriant	<u>≥</u> 50%	-	green w/ yellow back- ground
Enterococcus faecalis (29212) (00087*)	≥10 ³	inhibited	0%	_	_

Staphylococcus	≥10³	inhibited	0%	_	_
aureus (25923)					
(00034*)					

Key: * = corresponding WDCM Numbers

Storage and Shelf-life

Store between 2-8°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

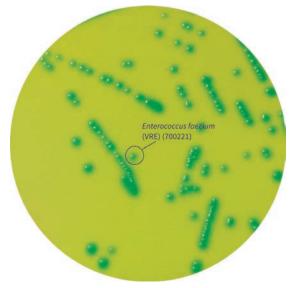
Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 4).

References

- Mara D., Horan NJ: The Handbook of water, wastewater and microbiology, Amsterdam,
 - The Netherlands, Academic Press; 2003.
- Mascini EM, Bonten MJ: Vancomycin- resistant enterococci: consequences for therapy and infection control. Clin Microbiol Infect.2005,11 (Suppl.4):43-56
- 3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
- Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.



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