



## Rapid HiEnterococci™ Agar

M1414

### Intended use

Recommended for rapid and easy identification and differentiation of Enterococci from water samples.

### Composition\*\*

Ingredients	g / L
Peptone, special	10.000
Sodium chloride	5.000
Sodium azide	0.300
Chromogenic mixture	0.060
Polysorbate 80 (Tween 80)	2.000
Disodium hydrogen phosphate	1.250
Agar	15.000
Final pH ( at 25°C)	7.5±0.2

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 33.61 gram in 1000 ml purified/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. Mix well and pour into sterile Petri plates.

### Principle And Interpretation

Enterococci are commonly found in the faeces of humans and other warm-blooded animals. Although some strains are ubiquitous and not related to faecal pollution, the presence of Enterococci in water is an indication of faecal pollution and the possible presence of enteric pathogens. The Enterococci test is recommended as a measure of ambient fresh and marine recreational water quality. Epidemiological studies have lead to the development of criteria which can be used to promulgate recreational water standards based on established relationships between health effects and water quality. The significance of finding Enterococci in recreational fresh or marine water samples is the direct relationship between the density of Enterococci and the risk of gastrointestinal illness associated with swimming in water (1,2). The Rapid HiEnterococci™ Agar allows for rapid identification and differentiation of Enterococci from water samples.

The peptone special supplies nitrogenous, carbonaceous compounds, long chain amino acids, vitamins and other essential nutrients. sodium chloride provides the osmotic balance for rapid growth of Enterococci. Sodium azide inhibits the accompanying microflora, especially the gram-negative organisms. The enzyme β-D-glucosidase present in Enterococci cleaves the chromogenic substrate, resulting in a blue green colour of the colonies.

### Type of specimen

Water samples

### Specimen Collection and Handling:

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

### Warning and Precautions :

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 specimens. Safety guidelines may be referred in individual safety data sheets

### Limitations

1. Some species may show poor growth due to nutritional variations.
2. Slight colour variation may be observed depending upon the utilization of the substrate by the organism.
3. Further biochemical and serological test are need to be carried out for confirmation.

## Performance and Evaluation

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

## Quality Control

### Appearance

Cream to yellow homogeneous free flowing powder

### Gelling

Firm, comparable with 1.5% Agar gel

### Colour and Clarity of prepared medium

Light amber coloured, clear to slightly opalescent gel forms in Petri plates

### Reaction

Reaction of 3.36% w/v aqueous solution at 25°C. pH : 7.5±0.2

### pH

7.30-7.70

### Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours

Organism	Inoculum (CFU)	Growth	Recovery	Colour of Colony
<i>Escherichia coli</i> ATCC 25922 (00013*)	50-100	none to poor	≤10%	
<i>Enterococcus faecalis</i> ATCC 29212 (00087*)	50-100	good	40-50%	blue green
<i>Pseudomonas aeruginosa</i> ATCC 27853 (00025*)	50-100	none to poor	≤10%	
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*)	50-100	good	40-50%	colourless

Key : (\*) Corresponding WDCM numbers.

## Storage and Shelf Life

Store between 15-25°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 in order 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. 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 sample must be decontaminated and disposed of in accordance with current laboratory techniques (4,5).

## Reference

1. Litsky W., Mallmann W. L., a Fifield C. W., 1953, Am. J. Pbl. Hlth.,43:873.
2. Manafi M., Sommer R., 1993, Wat. Sci. Tech. 27:271-274.
3. Lipps WC, Braun-Howland EB, Baxter TE,eds. Standard methods for the Examination of Water and Wastewater, 24th ed. Washington DC:APHA Press; 2023.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2<sup>nd</sup> Edition.
5. 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.Clinic

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