

## Technical Information

### Enterococcus Confirmatory MiVeg Agar

#### Product Code : VM1392

**Application:-** Enterococcus Confirmatory MiVeg Agar is recommended for confirming the presence of *Enterococci* in water supplies and other sources.

#### Composition

Ingredients	Gms / Litre
MiVeg hydrolysate	5.00
Yeast extract	5.00
Dextrose	5.00
Sodium azide	0.40
Methylene blue	0.01
Agar	15.00
Final pH (at 25°C)	8.0± 0.2

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

#### Principle & Interpretation

Enterococcus Confirmatory MiVeg Agar is prepared by adding MiVeg hydrolysate in place of Casein enzymic hydrolysate thus making the medium free from BSE/TSE risks. This media is the modification of the media devised by Sandholzer and Winter (1) for the detection of *Enterococci* in water supplies, swimming pools, sewage etc. The enterococci portion of the fecal *Streptococcus* group is a valuable indicator for determining the extent of fecal contamination of recreational surface waters (2).

MiVeg hydrolysate, Yeast extract, Dextrose provide necessary growth nutrients for *Enterococci*. Sodium azide inhibits gram-negative organisms. The positive presumptive tests are confirmed by inoculating Enterococcus Presumptive MiVeg Broth (VM1419) to Enterococcus Confirmatory slant-broth combination prepared with an azide agar medium (Enterococcus Confirmatory MiVeg Agar, VM1392) overlaid with a Salt Azide Penicillin MiVeg Broth (Enterococcus Confirmatory MiVeg Broth, VM1394). A negative catalase test is considered confirmed positive evidence of the presence of *Enterococci*. Single strength medium can be used for small inoculum. Production of acid and turbidity in an azide presumptive broth when incubated at 45°C is considered positive presumptive evidence for the presence of *Enterococci* which is confirmed by inoculating on Confirmatory Agar (VM1392).

#### Methodology

Suspend 30.4 grams of powder media in 1000 ml distilled water. Mix thoroughly. Heat to boiling to dissolve the medium completely. Dispense in 100 ml quantities in tubes and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Allow the agar tubes to cool in a slanted position.

**Warning :** Sodium azide has a tendency to form explosive metal azides with plumbing materials. It is advisable to use enough water to flush off the disposables

#### Quality Control

##### Physical Appearance

Yellow coloured, may have slightly greenish tinge, homogeneous, free flowing powder.

##### Gelling

Firm, comparable with 1.5% of Agar gel.

##### Colour and Clarity of prepared medium

Yellow coloured, slightly opalescent gel forms in tubes as slants.

**Reaction**

Reaction of 3.04% w/v aqueous solution is pH  $8.0 \pm 0.2$  at 25°C.

**pH Range**

7.8 – 8.2

**Cultural Response/Characteristics**

Cultural characteristics observed after an incubation at 45°C for 18 – 24 hours.

Organisms (ATCC)	Inoculum (CFU)	Growth	Recovery	Acid
<i>Enterococcus faecalis</i> (29212)	20-60	luxuriant	>70%	+
<i>Escherichia coli</i> (25922)	$10^3$	inhibited	0%	–

Key : + = acid production, yellow colour

– = no acid production, no colour change.

## Storage and Shelf Life

**Dried Media:** Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.

**Prepared Media:** 2-8° in sealable plastic bags for 2-5 day.

## Further Reading

1. Sandholzer and Winter, 1946, Commercial Fisheries Leaflet T1a.
2. Eaton A.D., Clesceie L.S. and Greebey A.E., (Eds.), 2005 Standard Methods for the Examination of Water and Wastewater, 21<sup>st</sup> Ed., APHA, Washington, D.C.
3. Splittstoesser, Wright and Hucker, 1961, Appl. Microbiol 9(4):303.

## Disclaimer :

- User must ensure suitability of the product(s) in their application prior to use.
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