

## Technical Information

### Enterococcus Confirmatory MiVeg Broth

#### Product Code : VM1394

**Application:-** Enterococcus Confirmatory MiVeg Broth 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
Sodium chloride	65.00
Final pH (at 25°C)	8.0 ± 0.2

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

#### Principle & Interpretation

Enterococcus Confirmatory MiVeg Broth 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 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). The Enterococcus Confirmatory MiVeg Broth has same formula as Enterococcus Confirmatory MiVeg Agar except agar, sodium chloride and Penicillin which is used to detect *Enterococci* from crab meat and oysters etc.

MiVeg hydrolysate, Yeast extract, Dextrose in the medium provide necessary growth nutrients for *Enterococci*. Sodium azide inhibits gram-negative organisms. Penicillin has inhibitory effect on *Staphylococci*. High concentration of NaCl (6.5%) serves selective enrichment of *Enterococcus*. The positive presumptive tests are confirmed by inoculating from 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 in Confirmatory Broth (VM1394).

#### Methodology

Suspend 80.4 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. Cool to room temperature and add 65 units of Penicillin to each 100 ml of broth prior to use.

**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.

### Colour and Clarity of prepared medium

Yellow coloured, clear solution which acquires greenish tinge at the surface on standing.

### Reaction

Reaction of 8.04% 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 24 hours.

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