Technical Information

Micro Vitamin Test Inoculum Broth

Product Code: DM 1133

Application: Micro Vitamin Test Inoculum Broth is recommended for preparation of inocula of used in microbiological *Lactobacilli* assays of vitamins.

Composition**

Ingredients	Gms / Litre
Yeast extract	20.000
Proteose peptone	5.000
Dextrose	10.000
Monopotassium phosphate	2.000
Polysorbate 80	0.100
Final pH (at 25°C)	6.7±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

Lactobacillus gram-positive, facultative anaerobic, lactic acid bacteria are so named because most of its members convert lactose and other sugars to lactic acid. They are usually benign in nature. How ever many species are play important role in decaying plant material. The production of lactic acid makes environment acidic which inhibits the growth of some harmful bacteria. Three types of media are generally used in microbiological assays namely (a) maintenance media, (b) inoculum /cultivation media and (c) the test assay media.

Micro Vitamin Test Inoculum Broth is used for maintaing stock cultures of Lactobacilli and other test organisms being used in microbiological assays [(1)]. This media can also be used for routine cultivation of *Lactobacilli* and inoculum preparation in microbiological assays of vitamins.

Proteose peptone and yeast extract in the medium provide nitrogen, sulphur, vitamins and other essential nutrients for growth. Dextrose is the energy source. Polysorbate 80 is the fatty acid source. Monopotassium phosphate buffers the medium. Stock cultures are prepared by stab inoculation in triplicates. One is used for preparation of stock cultures while others are used for inoculum preparation for assays. Transfer of cultures should be done at weekly or biweekly intervals. For this suspend a 16-24 hours culture of Lactobacilli from Micro Vitamin Test Culture Agar into Micro Vitamin Test Inoculum Broth. After an incubation at 35-37°C for 18-24 hours, centrifuge the culture and decant the supernatant. Re-suspend the deposit in 10 ml of sterile saline suspension. Repeat the washing two more times. Dilute the washed cell suspension with basal assay medium to obtain the required cells density. For procedure of Vitamin Assay, refer standard references [(2)].

Methodology

Suspend 37.1 grams of powder media in 1000 ml distilled water. Shake well & heat if necessary to dissolve the medium completely. Dispense and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Quality Control

Physical Appearance
Cream to yellow homogeneous free flowing powder
Colour and Clarity of prepared medium
Light yellow coloured clear solution in tubes
Reaction
Reaction of 3.7 1% w/v aqueous solution at 25°C. pH : 6.7±0.2
pH range 6.50-6.90

Cultural Response/Characteristics

DM1133: Cultural characteristics observed after an incubation at 35-37^{0}C for 24-48 hours.

Organism	Inoculum (CFU)	Growth
Lactobacillus casei ATCC 9595	50-100	Good-luxuriant
Lactobacillus leichmanni ATCC 7830	50-100	Good-luxuriant
Lactobacillus plantarum ATCC 8014	50-100	Good-luxuriant
Lactobacillus viridescens ATCC 12706	50-100	Good-luxuriant

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: 2-8^0 in sealable plastic bags for 2-5 days.

Further Reading

1. Atlas R. M., 1993, Handbook of Microbiological Media, Parks L.C., (Ed.), CRC Press, Inc.
2. Horwitz, (Ed.), 2000, Official Methods of Analysis of AOAC International, 17th Ed., Vol. I, AOAC International, Gaithersburg, MD.

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