



Dehydrated Culture Media
Bases / Media Supplements

Technical Information

B12 Inoculum Broth (*L. leichmannii*)

Product Code: DM 1206

Application: B12 Inoculum Broth is used for preparing the inoculum of *Lactobacillus leichmannii* ATCC 7830 in the microbiological assay of Vitamin B12.

Composition**

Ingredients	Gms / Litre
Proteose peptone	7.500
Yeast extract	7.500
Dextrose	10.000
Polysorbate 80	0.100
Monopotassium phosphate	2.000
Tomato juice (from 100 ml)	5.000
Final pH (at 25°C)	6.8±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

This medium which is rich in nutrients, is recommended by USP for inoculum preparation of *Lactobacillus leichmannii* ATCC 7830, being used in microbiological estimation of Vitamin B12. *Lactobacillus* species have very exacting nutritional requirements for amino acids and vitamins. This restricts them to nutritionally compete in the environment. *Lactobacillus* species grow poorly on non-selective media. Kulp⁽²⁾ found that the growth of *Lactobacillus acidophilus* was enhanced with the addition of tomato juice, which was similar to the finding of Mickle and Breed⁽³⁾ Proteose peptone serves as a source of nitrogen and amino acids. Yeast extract is the vitamin source. Tomato juice is added to create the proper acidic environment. Dextrose is the carbon source and Polysorbate 80 acts as an emulsifier. Monopotassium phosphate provides buffering capacity. For preparing inoculum, the culture is grown in 5 ml sterile B12 Inoculum Broth for 18 to 24 hours at 35°C and then the culture is centrifuged to obtain cell sediment. The supernatant is decanted and the cells sediment is suspended in B12 Assay Medium (DM1036). This cell suspension is used as an inoculum after adjusting its density.

Methodology

Suspend 32.1 grams of powder media in 1000 ml distilled water. Shake well and heat if necessary to dissolve the medium completely. Distribute in tubes 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

Amber coloured, clear to slightly opalescent solution in tubes

Reaction

Reaction of 3.2% w/v aqueous solution at 25°C. pH : 6.8±0.2

pH Range

6.60-7.00





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Cultural Response/Characteristics

DM1206: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth
<i>Lactobacillus leichmannii</i> ATCC 7830	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: 2-8⁰ in sealable plastic bags for 2-5 days.

Further Reading

1. The United States Pharmacopoeia, 2006, USP 29/ NF 24, The United States Pharmacopoeial Convention, Rockville, MD.
2. Kulp and White, 1932, Science 76:17.
3. Mickle and Breed, 1925, Technical Bulletin 110, NY State Agriculture Ex. station, Geneva, N. Y.

Disclaimer :

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