TM 1868 - VITAMIN B12 AGAR

INTENDED USE

For microbiological assay of Vitamin B12 by using *Lactobacillus leichmannii* as test organism.

PRODUCT SUMMARY AND EXPLANATION

Incorporation of Vitamin B12 in specified increasing amounts gives a growth response that can be measured by the diameter of the zone of growth around the disc or cup containing Vitamin B12.

Inoculum for the assay is prepared by sub culturing from a stock culture previously made by stab inoculation. Freshly subcultured organisms incubated at 37°C for 24 hours, centrifuged, washed and suspended in 10 ml saline are recommended for the assay. The growth response obtained is turbid metrically or acidimetrically measured.

A standard curve is plotted with absorbance as a function of the vitamin B12 concentration. The concentration of vitamin B12 in the test sample is calculated based on the interpretation of the standard curve.

Extreme care should be taken to avoid contamination of media or glassware used for the assay. Detergent-free clean glassware should be used. Even small amount of contamination by foreign material may lead to erroneous results.

The test organism used for inoculating must be cultured and maintained on media recommended for this purpose.

COMPOSITION

Ingredients	Gms / Ltr
Casein acid hydrolysate, vitamin free	10.000
Soyapeptone, vitamin free	5.000
Dextrose	20.000
Sodium acetate	12.000
Polysorbate 80	1.000
Potassium sulphate	20.000
Monopotassium phosphate	1.000
Dipotassium phosphate	1.000
Magnesium sulphate	0.400
Sodium chloride	0.020
Ferrous sulphate	0.020
Manganese sulphate	0.020
Ribonucleic acid	1.000
Sodium thioglycollate	1.700
L-Cystine	0.200
Adenine sulphate	0.0176
Guanine hydrochloride	0.0124
Uracil	0.010
Xanthine (sodium)	0.010
Folic acid	0.001
Riboflavin (Vitamin B2)	0.002
Thiamine hydrochloride	0.002

Calcium pantothenate	0.002
Niacin	0.002
Pyridoxine hydrochloride	0.004
Pyridoxal 5 phosphate	0.004
Biotin	0.000001
DL-Tryptophan	0.200
Agar	15.000

PRINCIPLE

Vitamin B12 Agar is a dehydrated medium devoid of Vitamin B12 containing all the nutrients essential for the growth of *Lactobacillus leichmannii.*

INSTRUCTION FOR USE

- Dissolve 88.62 grams in 1000 ml distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes.
- Mix well and pour into sterile Petri plates.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Off-white to yellow homogeneous powder having a tendency to form soft lumps, which can be easily broken down to powder form.

Appearance of prepared medium : Light amber coloured clear to slightly opalescent gel forms in Petri plates.

pH (at 25°C) : 6.2±0.2

INTERPRETATION

Cultural characteristics observed after an incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Incubation Temperature	Incubation Period
Lactobacillus leichmanii	4797	50-100	Good	40-50%	35-37°C	18-24 Hours

PACKAGING:

In pack size of 100 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 2-8°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

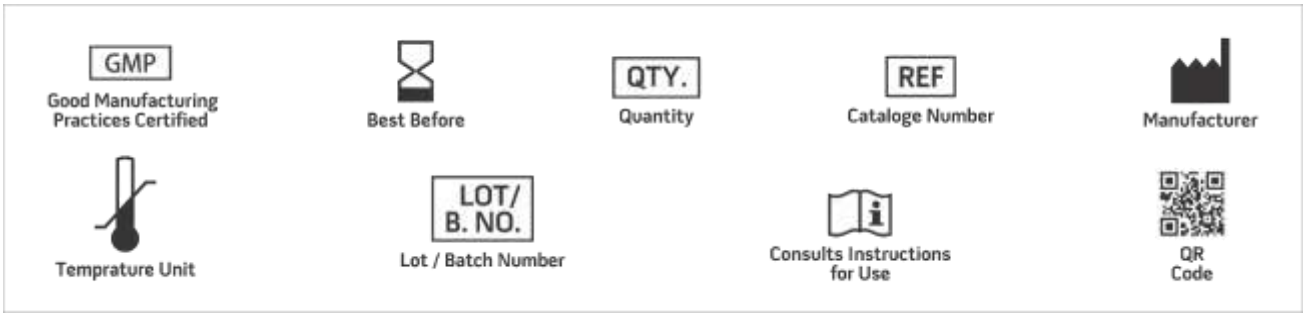
Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

1. The United States Pharmacopoeia, 2006, USP29/NF24, The United States Pharmacopeial Convention, Rockville, MD.
2. H. Williams, (Ed.), 2005, Official Methods of Analysis of the Association of Official Analytical Chemists, 19th Ed., AOAC, Washington, D.C



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

***For Lab Use Only**

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