

PRODUCT SPECIFICATION SHEET

Micro Vitamin Test Culture Agar (DM165)

Intended Use

Micro Vitamin Test Culture Agar (DM165) is recommended for cultivation and maintenance of stock cultures of *Lactobacilli* used in vitamin assays

Product Summary and Explanation

Lactobacillus is a genus of gram-positive facultative anaerobic or microaerophilic rod-shaped bacteria. They are a major part of the lactic acid bacteria group. They are common and usually benign. Many species are prominent in decaying plant material. The lactic acid bacteria are so named because most of its members convert lactose and other sugars to lactic acid. Production of lactic acid makes its environment acidic which inhibits the growth of some harmful bacteria. *Lactobacillus* species grow poorly on non-selective culture media and require special nutrients. Generally three types of media are used in microbiological assays namely maintenance media, inoculum /cultivation media and the test assay media.

Micro Vitamin Test Agar is used for carrying stock cultures of *Lactobacilli* and other test organisms used in microbiological assays.⁽¹⁾ The routine cultivation of *Lactobacilli* in microbiological assays of vitamins and in inoculum preparation for assays can also be done using this medium.

Principles of the Procedure

Micro Vitamin Test Culture Agar contains peptic digest of animal tissue and yeast extract which provide nitrogen, sulphur, vitamins and other essential nutrients required for growth of organisms. Dextrose is the carbon and energy source. Polysorbate 80 is the fatty acid source. Monopotassium phosphate acts as a buffering agent.

Formula / Liter

Ingredients	Gms / Liter
Yeast extract	20.00
Peptic digest of animal tissue	5.00
Dextrose	10.00
Monopotassium phosphate	2.00
Polysorbate 80	0.10
Agar	15.00
Final pH: 6.7 ± 0.2 at 25°C	
Formula may be adjusted and/or supplemented as required to meet performance specifications	

Precautions

1. For Laboratory Use only.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

Directions

1. Suspend 52.1 grams of the medium in one liter of distilled water.
2. Heat to boiling, to dissolve the medium completely.
3. Dispense and autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.

Quality Control Specifications

Dehydrated Appearance	Cream to yellow homogeneous free flowing powder
Prepared Medium	Light yellow coloured clear to slightly opalescent gel forms in tubes as butts
Reaction of 5.2% Solution	pH : 6.7 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 1.5% Agar gel

Expected Cultural Response: Cultural characteristics observed after an incubation at 35 - 37°C for 24 - 48 hours.

Sr. No.	Organisms	Results to be achieved	
		Inoculum (CFU)	Growth



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1.	<i>Lactobacillus casei</i> ATCC 9595	50 -100	good-luxuriant
2.	<i>Lactobacillus viridescens</i> ATCC 12706	50 -100	good-luxuriant
3.	<i>Lactobacillus leichmanni</i> ATCC 4797	50 -100	good-luxuriant
4.	<i>Lactobacillus plantarum</i> ATCC 8014	50 -100	good-luxuriant

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure

1. 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 made at weekly or biweekly intervals.
2. Suspend a 16-24 hours culture of from Micro Vitamin Test Culture Agar into Micro Vitamin Test Inoculum Broth.
3. After an incubation at 35-37°C for 18-24 hours, centrifuge the culture and decant the supernatant. Re-suspend the centrifuged cells in 10 ml of sterile saline suspension. Repeat the washing two more times. Dilute the washed cell suspension with basal assay medium or as desired to obtain the required density of cells.
4. For procedure of Vitamin Assay, refer standard references.⁽²⁾

Results

Refer appropriate references and test procedures for interpretation of results.

Storage

Store the sealed bottle containing the dehydrated medium at 10 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light.

Expiration

Refer to the expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedure

1. For identification, organisms must be in pure culture. Morphological, biochemical and/or serological tests should be performed for final identification.
2. Consult appropriate texts for detailed information and recommended procedures.

Packaging

Product Name : Micro Vitamin Test Culture Agar

Product Code : DM 165

Available Pack sizes : 100gm

References

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.

Further Information

For further information please contact your local MICROMASTER Representative.



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DM165PSS,QAD/FR/024,Rev.00

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