

Soyabean casein Digest Agar w/ Lecithin and Polysorbate 80 Tryptone Soya Agar w/ Lecithin and Polysorbate 80 (Microbial Content Test Agar) Intended use

M449

Recommended for determining efficiency of sanitization of containers, equipment surfaces, water miscible cosmetics etc.

Composition**	
Ingredients	Gms / Litre
Tryptone	15.000
Soya Peptone	5.000
Sodium chloride	5.000
Lecithin	0.700
Polysorbate 80 (Tween 80)	5.000
Agar	15.000
Final pH (at 25°C)	7.3±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 45.7 grams in 1000 ml purified/distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Tryptone Soya Agar with Lecithin and Polysorbate 80 is used in RODAC (Replicate Organism Detection and Counting) plates (1) for the detection and enumeration of microorganisms present on surfaces of sanitary importances (2,3). Tryptone and Soya peptone provide nitrogenous compounds and other nutrients essential for microbial replication. Lecithin and polysorbate 80 (Tween 80) are neutralizers reported to inactivate residual disinfectants from where the sample is collected (4). Lecithin neutralizes quaternary ammonium compounds and polysorbate 80 neutralizes phenolic disinfectants, hexachlorophene, formalin and with lecithin ethanol (5).

Collection of samples from areas before and after the treatment with disinfectant evaluates cleaning procedures in environmental sanitation. The presence and number of microorganisms is determined by the appearance of colonies on the agar surface (6). After counting the colonies, carry out biochemical testing for identification.

Type of specimen

Pharmaceutical samples, Swabs of containers, Equipment surfaces, Water miscible cosmetics etc.

Specimen Collection and Handling

For swabs of containers, equipment surfaces, water miscible cosmetics samples follow appropriate techniques for handling specimens as per established guidelines (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

- 1. This medium is general purpose medium and may not support the growth of fastidious organisms.
- 2. Further biochemical and serological test must be carried out for complete identification.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light yellow to medium amber coloured clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of 4.57% w/v aqueous solution at 25°C. pH : 7.3±0.2

pН

7.10-7.50

Cultural Response

Growth Promotion was observed after an incubation at 30-35°C for 18-24 hours for bacteria and for fungus <=5 days.

Recovery rate

Recovery rate is considered 100% for bacterial growth on : Soyabean Casein Digest Agar and fungal growth on Sabouraud Dextrose Agar.

Growth promoting properties

Growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating ≤ 100 cfu (at 30-35°C for ≤ 18 hours).

Neutralization Test

The smaller zone of inhibition compared to SCDA indicates neutralization of quaternary ammonium compounds by this medium.

Organism	Inoculum (CFU)	Growth	Observed Lot value (CFU)	Recovery	Incubation temperature	Incubation period
Growth promoting						
**Bacillus spizizenii ATCC 6633 (00003*)	50 -100	luxuriant	35 -100	>=70 %	30 -35 °C	18 -24 hrs
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*)	50 -100	luxuriant	35 -100	>=70 %	30 -35 °C	18 -24 hrs
Staphylococcus aureus subsp. aureus ATCC 6538 (00032*)	50 -100	luxuriant	35 -100	>=70 %	30 -35 °С	18 -24 hrs
<i>Escherichia coli</i> ATCC 25922 (00013*)	50 -100	luxuriant	35 -100	>=70 %	30 -35 °C	18 -24 hrs
<i>Escherichia coli</i> ATCC 8739 (00012*)	50 -100	luxuriant	35 -100	>=70 %	30 -35 °C	18 -24 hrs
Pseudomonas aeruginosa ATCC 27853 (00025*)	50 -100	luxuriant	35 -100	>=70 %	30 -35 °C	18 -24 hrs
^Pseudomonas paraeruginosa ATCC 9027 (00026*)	50 -100	luxuriant	35 -1	>=70 %	30 -35 °С	18 -24 hrs
Salmonella Abony NCTC 6017	50 -100	luxuriant	35 -100	>=70 %	30 -35 °С	18 -24 hrs
<i>\$ Kokuria rhizophila</i> ATCC 9341	50 -100	luxuriant	35 -100	>=70 %	30 -35 °C	18 -24 hrs
Salmonella Typhimurium ATCC 14028 (00031*)	50 -100	luxuriant	35 -100	>=70 %	30 -35 °C	18 -24 hrs
Candida albicans ATCC 10231 (00054*)	50 -100	luxuriant	35-100	>=70 %	30 -35 °C	<=5 d
Candida albicans ATCC 2091 (00055*)	50 -100	luxuriant	35 -100	>=70 %	30 -35 °C	<=5 d
#Aspergillus brasiliensis ATCC 16404 (00053*)	50 -100	luxuriant	35 -100	>=70 %	30 -35 °C	<=5 d
<i>Candida albicans</i> ATCC 10231 (00054*)	50 -100	luxuriant	35 -100	>=70 %	20 -25 °C	<=5 d

Candida albicans ATCC 2091 (00055*)	50 -100	luxuriant	35 -100	>=70 %	20 -25 °C	<=5 d
#Aspergillus brasiliensis ATCC 16404 (00053*)	50 -100	luxuriant	35 100	>=70 %	20 -25 °C	<=5 d
Clostridium sporogenes ATCC 19404 (00008*)	50 -100	luxuriant	35-100	>=70 %	30 -35 °C	<=48 hours

Key : (*) Corresponding WDCM numbers.

[^] Formerly known as *Pseudomonas aeruginosa*

**Formerly known as *Bacillus subtilis* subsp. *spizizenii* \$ Formerly known as *Micrococcus luteus*

Formerly known as Aspergillus niger

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (7,8).

Reference

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6. Murray PR, Baron, Pfaller, and Yolken (Eds.), 2003, In Manual of Clinical Microbiology, 8th ed., ASM, Washington, D.C.

7. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.

8. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

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