

Tryptic Soy Agar with Lecithin and Polysorbate 80

INTENDED USE

Tryptic Soy Agar with Lecithin and Polysorbate 80 is used for the quantitation of viable microbial contaminants from surfaces and equipment in controlled environments.

SUMMARY AND EXPLANATION

Microbial monitoring programs for controlled environments assess the effectiveness of cleaning and sanitation practices by personnel.¹⁻⁷ Routine testing should include microbial counts of room air, compressor air entering the critical area, surfaces, equipment, sanitation containers, floors, walls, and personnel garments.

For examination of surfaces in critical environments, sampling is accomplished by use of contact plates or by the swabbing method.¹⁻⁷ Contact plates are used on flat surfaces such as walls, floors, ceilings and some equipment surfaces, then directly incubated at the appropriate time and temperature for quantitation of viable counts. The swabbing method is used for irregular surfaces with cracks, corners or crevices. The swab is then placed in a diluent and an estimate of the microbial count is done by plating an aliquot on or in a specified agar. The area to be swabbed is defined using a sterile template of appropriate size, usually 24 to 30 cm². The microbial estimates are reported per contact plate or per swab.

A commonly used all-purpose medium used for sampling surfaces in a controlled area is Tryptic Soy Agar (Soybean-Casein Digest Agar).¹ When disinfectants or antibiotics are used in the controlled area, media with the appropriate inactivating agents are required.¹⁻⁷ For this purpose, 0.7 g of lecithin and 5.0 g of Polysorbate 80 may be added to one liter of agar, before sterilization.^{2,6}

Tryptic Soy Agar with Lecithin and Polysorbate 80 is recommended in Chapter <797> – Pharmaceutical Compounding – Sterile Preparations and Chapter <1116> – Microbiological Evaluation of Clean Rooms of the *USP* for use when sampling surfaces for viable microbial contaminants in controlled environments.¹ The formulation is available as a prepared plate in various configurations, such as a contact plate, a 100 mm standard plate and a 150 mm settling plate. The product is also available in a 100 mL and 500 mL pour bottle and a 20 mL pour tube (please refer to the Content section at the end of this package insert).

Tryptic Soy Agar meets United States Pharmacopeia (USP) performance specifications.

PRINCIPLE OF THE TEST

Tryptic Soy Agar with Lecithin and Polysorbate 80 contains enzymatic digests of casein and soybean meal that are excellent sources of nitrogen, amino acids, and peptides. Sodium chloride provides electrolytes to help maintain osmotic balance of the medium. Agar is the solidifying agent. Lecithin is incorporated to neutralize quaternary ammonium compounds.¹ Polysorbate 80 neutralizes hexachlorophene, formalin, and phenols.⁸

COMPOSITION

Approximate*, per liter of purified water.

Tryptic Soy Agar with Lecithin and Polysorbate 80

Enzymatic Digest of Casein	15.0 g
Enzymatic Digest of Soybean Meal	5.0 g
Sodium Chloride	5.0 g
Lecithin	0.7 g
Polysorbate 80	5.0 g
Agar	20.5 g

Final pH 7.3 ± 0.2 at 25°C

*Adjusted and/or supplemented to meet performance standards.

ADDITIONAL MATERIALS REQUIRED BUT NOT PROVIDED

Standard supplies and equipment commonly found in a microbiology laboratory are not provided.

STORAGE INSTRUCTIONS

Upon receipt, store plates at 2-8°C away from direct light in an inverted position. Store bottles according to the package label in an upright position. Media should not be used if there are signs of contamination, deterioration (i.e., shrinking, cracking or discoloration), or if the expiration date has passed. Store plates in original outer wrapping until ready to use.

Media can be inoculated up to the expiration date and incubated for the appropriate incubation period.

WARNINGS AND PRECAUTIONS

1. For laboratory use only.
2. Observe approved biohazard precautions. All samples, microbial cultures, and inoculated products should be considered infectious and handled appropriately.
3. Aseptic technique and usual precautions for handling the bacterial group studied should be observed throughout this procedure. Refer to "CLSI® M29-A3, *Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline* – Third Edition."⁹
4. For additional information on handling precautions, refer to the latest edition of *Biosafety in Microbiological and Biomedical Laboratories*,¹⁰ or the current regulations in the country of use.

This culture medium should not be used as manufacturing material or as a component of a drug. The medium should be used according to the procedure indicated in this package insert. Any change or modification in the procedure may affect the results.

SAMPLE COLLECTION AND VALIDATION

Samples should be collected using appropriate techniques, protected from extreme heat and cold, and delivered to the laboratory without delay. Follow procedures currently recommended in standard methods for collection of samples when testing surfaces in controlled environments.¹⁻⁷

INSTRUCTIONS FOR USE

1. Prior to inoculation, the media should be brought to room temperature. The agar surface should be flat and moist without excessive moisture.
2. Refer to appropriate references for details on test methods using Tryptic Soy Agar with Lecithin and Polysorbate 80.¹⁻⁷
3. *For melting bottled or tubed media*, loosen caps and place in boiling water bath (100°C) until melted. The water level in the bath should reach the media line. Once melted, cool media to 45-50°C before use.

NOTE: Use of a microwave oven to melt tubed and bottled media is not recommended.

READING AND INTERPRETATION

Refer to appropriate references for details on plate count methods.¹¹⁻¹³

QUALITY CONTROL**Prepared Appearance**

Light to medium amber, clear to trace hazy

Cultural Response

Inoculate plates in duplicate and incubate aerobically as directed below.

Microorganism	ATCC®	Inoculum CFU	Incubation Conditions at 30-35°C	Expected Results with > 50% recovery
<i>Staphylococcus aureus</i>	6538™	< 100	18-72 hours at 30-35°C	Growth
<i>Bacillus subtilis</i>	6633™	< 100	18-72 hours	Growth
<i>Escherichia coli</i>	8739™	< 100	18-72 hours	Growth
<i>Pseudomonas aeruginosa</i>	9027™	< 100	18-72 hours	Growth
<i>Candida albicans</i>	10231™	< 100	2-5 days	Growth
<i>Aspergillus brasiliensis</i>	16404™	< 100	2-5 days	Growth

LIMITATIONS OF THE TEST

- For identification, organisms must be in pure culture. Perform biochemical/serological tests for complete identification. Consult appropriate references for further information.¹¹⁻¹³
- Accurate counts may be difficult when molds or spreading colonies are present.

WASTE DISPOSAL







Unused products may be considered as non-hazardous waste and disposed of accordingly.

Dispose of all used products as well as any other contaminated disposable materials following procedures for infectious or potentially infectious products. It is the responsibility of each laboratory to handle waste and effluents produced according to their nature and degree of hazardousness and to treat and dispose of them (or have them treated and disposed of) in accordance with any applicable regulations.

REFERENCES

- United States Pharmacopeial Convention, Inc. The United States Pharmacopeia 34/The National Formulary 29, Suppl. 1 [online]. Rockville, MD; 2011.
- Dyer RL, Frank JF, Johnson B, Hickey P. Microbiological tests for equipment, containers, water and air. *Standard methods for the examination of dairy products*, 17th ed. Washington, DC: American Public Health Association; 2004:333-335.
- Greger G. Basic Requirements for Aseptic Manufacturing of Sterile Medicinal Products - A Comparison Between Europe and USA. 2004. US Department of Health and Human Services, Food and Drug Administration. *Guidance for industry: Sterile drug products produced by aseptic processing – current good manufacturing practice*; 2004.
- Parenteral Drug Association. PDA Technical Report No. 13 (Revised). Fundamentals of an environmental monitoring program. *PDA J. Pharm. Sci. Technol.* 2001;55(5 Suppl TR13):1-35.
- Downes FP, Ito K (ed.). *Compendium of methods for the microbiological examination of foods*, 4th ed. Washington, DC: American Public Health Association; 2001.
- International Organization for Standardization. *Cleanrooms and associated controlled environments – Biocontamination control – Part 1: General principles and methods*. ISO 14698-1. First ed. Geneva, Switzerland: International Organization of Standardization; 2003.
- Kastango ES, Faylor K. The importance of environmental monitoring, Part II: Surface testing. *Pharmacy Purchasing & Products*. 2005;2:24-26.
- Clinical and Laboratory Standards Institute. *Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline – M29-A3*. Wayne, PA: CLSI; 2005.
- US Department of Health and Human Services, Centers for Disease Control and Prevention and National Institutes of Health. *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*. 5th Edition. Washington, DC: US Government Printing Office; 2007.
- Versalovic J, Carroll KC, Funke G, Jorgensen JH, Landry ML, Warnock DW (ed.). *Manual of clinical microbiology*. 10th ed. Washington, DC: American Society for Microbiology; 2011.
- Garcia LS. (ed.). *Clinical microbiology procedures handbook*. 3rd ed. Washington, DC: American Society for Microbiology; 2010.
- Forbes BA., Sahm DF, Weissfeld AS. *Bailey and Scott's diagnostic microbiology*. 12th ed. St. Louis, MO: Mosby, Inc; 2007.

INDEX OF SYMBOLS


Symbol	Meaning
	Catalogue number
	Legal manufacturer
	Temperature limitation
	Use by date
	Batch code
	Consult Instructions for Use

WARRANTY

bioMérieux warrants the performance of the product for its stated intended use provided that all procedures for usage, storage and handling, shelf life (when applicable), and precautions are strictly followed as detailed in the instructions for use (IFU).

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CONTENT

Prepared Media		
	Name	Units/Pkg
C6045	Tryptic Soy Agar with Lecithin and Polysorbate 80, FIX SYSTEM® Contact Plate, 60 mm, 14.6 mL	10
M1060	Tryptic Soy Agar with Lecithin and Polysorbate 80, 90 mm, 25 mL	10
P2810	Tryptic Soy Agar with Lecithin and Polysorbate 80, Pour bottle, 500 mL	10
T8100-C	Tryptic Soy Agar with Lecithin and Polysorbate 80, Pour tube, 20 x 150 mm, 20 mL	100

Instructions for Use provided in the kit or downloadable from www.biomerieux.com/techlib

REVISION TABLE

This section contains a summary of changes made to each released revision of this document starting with part number 9302249A.

Revision Date	Revision Number	Change Type	Change Summary
2011-12	9302249 A	N/A	Creation of new document.
2013-04	9305264 B	Content Change	Revisions to Composition section and IP footer.
2015-02	9309220 C	Content Change	Update fill volume and diameter for M1060 in Content section.
			Delete REF L2041 in Content section. Product is obsolete.
			Addition of web address for instructions for use access.
		Administrative	Update heading names to reflect current bioMérieux instructions for use template.
2016-10	9313342 D	Content Change	Formatting changes to reflect current bioMérieux instructions for use template.
			Removed obsolete product REF P2230 under Content, replaced previous warranty statement with current warranty language, added technical library link.

NOTE: Minor typographical, grammar, and formatting changes are not included in the revision history.

Change Type categories:

- **Correction** = Correction of documentation anomalies.
- **Content Change** = Implementation of new and modified (updated) intended use and performance characteristics.
- **Administrative** = Implementation of non-technical changes noticeable to the user.
- **N/A** = Not applicable

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