

Phenylethanol Agar (NCM0153)

Intended Use

Phenylethanol Agar is used with blood for the selective isolation of Gram-positive cocci in a laboratory setting. Phenylethanol Agar is not intended for use in the diagnosis of disease or other conditions in humans.

Description

Brewer and Lilley reported the addition of phenylethanol to a nutritive medium permitted growth of Gram-positive organisms, but markedly to completely inhibited growth of Gram-negative organisms. Phenylethanol Agar inhibits swarming of *Proteus* spp., and can be used to selectively isolate anaerobic bacteria from specimens with mixed flora. Phenylethanol Agar is specified for use in several reference methods.

Typical Formulation

Enzymatic Digest of Casein	15.0 g/L
Enzymatic Digest of Soybean Meal	5.0 g/L
Sodium Chloride	5.0 g/L
Phenylethanol	2.5 g/L
Agar	15.0 g/L

Final pH: 7.3 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precaution

Refer to SDS

Preparation

1. Suspend 42.5 g of the medium in one liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes.
4. Cool to 45 - 50°C.
5. Prepare 5 - 10% blood agar by aseptically adding the appropriate volume of sterile defibrinated blood to melted sterile agar medium.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, beige with soft lumps.

Prepared Appearance: Prepared medium is trace to slightly hazy and pale yellow. Prepared medium with 5% sheep blood is red and opaque.

Expected Cultural Response: Cultural response on Phenylethanol Agar at 35 ± 2°C and examined for growth after 18 - 48 hours incubation in an aerobic atmosphere.

Technical Specification Sheet



Microorganism	Approx. Inoculum (CFU)	Expected Growth Results
<i>Enterococcus faecalis</i> ATCC® 29212	10-300	Fair to good growth
<i>Escherichia coli</i> ATCC® 25922	1000	If recovered, pinpoint colonies
<i>Proteus mirabilis</i> ATCC® 12453	1000	If recovered, pinpoint colonies
<i>Staphylococcus aureus</i> ATCC® 25923	10-300	Fair to good growth
<i>Staphylococcus epidermidis</i> ATCC® 12228	10-300	Fair to good growth
<i>Streptococcus pneumoniae</i> ATCC® 6305	10-300	Fair to good growth
<i>Streptococcus pyogenes</i> ATCC® 19615	10-300	Fair to good growth

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

Test Procedure

1. Process each specimen as appropriate, inoculate directly onto surface of the medium. Streak for isolation with inoculating loop.
2. Incubate plates at 35°C under conditions of increased CO₂ (5 - 10%) for 18 - 24 hour, and if necessary, 40 - 48 hours.

Results

Examine medium for growth and hemolytic reactions after 18 - 24 and 48 hours incubation. Perform additional biochemical testing to identify the organism.

Expiration

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

Limitations of the Procedure

1. Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.
2. Some Gram-positive cocci may be slightly inhibited and many require further incubation (up to 48 hours) for sufficient growth to be evident.
3. Subculture Gram-positive colonies onto nonselective medium for biochemical testing.
4. When supplemented with blood this medium may demonstrate atypical hemolytic reactions. The prepared medium should not be used for the classification or determination of hemolytic reactions.
5. *Pseudomonas aeruginosa* is **not** inhibited on this medium.

Storage

Store dehydrated culture media at 2-8°C away from direct sunlight. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

References

1. Brewer, J. H., and B. D. Lilley. 1949. Paper presented at the December meeting of the Maryland Association of Medical and Public Health Laboratories.
2. Lilley, B. D., and J. H. Brewer. 1953. The selective antibacterial action of phenylethylalcohol. J. Pharm. Assoc. 42:6.
3. Murray, P. R., E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Tenover (eds.). Manual of clinical microbiology, 6th ed. American Society of Microbiology, Washington, D.C.
4. Isenberg, H. D. 1992. Clinical microbiology procedures handbook, American Society for Microbiology, Washington, D.C.



Technical Specification Sheet



5. Washington, J. A., Jr. 1981. Laboratory procedures in clinical microbiology. Springer-Verlag, New York.
6. MacFaddin, J. F. 1985. Media for the isolation-cultivation-identification-maintenance of medical bacteria, vol. 1 Williams & Wilkins, Baltimore, MD.

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