

D/E NEUTRALIZING AGAR (7375)

Intended Use

D/E Neutralizing Agar is used for the isolation of microorganisms from sanitized environmental surfaces.

Product Summary and Explanation

D/E Neutralizing Agar was developed by Dey and Engley¹ to neutralize a broad spectrum of disinfectants and preservative antimicrobial chemicals, including quaternary ammonium compounds, phenolics, iodine, chlorine preparations, mercurials, formaldehyde, and glutaraldehyde. D/E Neutralizing media neutralize higher concentrations of residual antimicrobials when compared with other standard neutralizing formulations, such as Lethen media, Thioglycollate media, and Neutralizing Buffer.^{2,3}

Total neutralization of disinfectants is critical. Disinfectant residues can result in a false negative (no-growth) test. D/E Neutralizing Agar effectively neutralize the inhibitory action of disinfectant carryover,^{4,5} allowing differentiation between bacteriostasis and true bactericidal action of disinfectant chemicals. This is a critical characteristic to consider when evaluating a disinfectant. D/E Neutralizing Agar is recommended for use in disinfectant evaluations, environmental sampling (swab and contact plate methods), and testing of water-miscible cosmetics.⁶

Principles of the Procedure

Enzymatic Digest of Casein and Yeast Extract provides nitrogen, carbon, vitamins, and minerals in D/E Neutralizing Agar. Dextrose is a source of fermentable carbohydrate. Sodium Thioglycollate neutralizes mercurials. Sodium Thiosulfate neutralizes iodine and chlorine. Sodium Bisulfite neutralizes formaldehyde and glutaraldehyde. Lecithin neutralizes quaternary ammonium compounds and Polysorbate 80 neutralizes phenols, hexachlorophene, formalin, and with lecithin, ethanol. Bromcresol Purple is used as a colorimetric indicator to demonstrate the production of acid from the fermentation of dextrose.

Formula / Liter

Enzymatic Digest of Casein.....	5 g
Yeast Extract	2.5 g
Dextrose.....	10 g
Sodium Thioglycollate.....	1 g
Sodium Thiosulfate	6 g
Sodium Bisulfite	2.5 g
Polysorbate 80.....	5 g
Lecithin (Soybean).....	7 g
Bromcresol Purple	0.02 g
Agar	15 g

Final pH: 7.6 ± 0.2 at 25°C

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

Precautions

1. For Laboratory Use.
2. HARMFUL. Harmful if inhaled, swallowed, or absorbed through the skin. May cause allergic respiratory reaction. Irritating to eyes, respiratory system, and skin.

Directions

1. Suspend 54 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.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, lumpy, and blue-gray to green.

Prepared Appearance: Prepared medium is purple and opaque.

Expected Cultural Response: Cultural response in D/E Neutralizing Agar incubated aerobically at 35 ±2°C and examined for growth after 40 - 48 hours.

Microorganism	Approx. Inoculum (CFU)	Expected Results
<i>Bacillus subtilis</i> ATCC® 9372	10 - 300	Growth
<i>Escherichia coli</i> ATCC® 25922	10 - 300	Growth
<i>Pseudomonas aeruginosa</i> ATCC® 27853	10 - 300	Growth
<i>Salmonella typhimurium</i> ATCC® 14028	10 - 300	Growth

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

Test Procedure

D/E Neutralizing Agar is used in a variety of procedures. Consult appropriate references for complete information.⁶

Results

Refer to appropriate references and procedures for results.

Storage

Store sealed bottle containing the dehydrated medium at 2 - 8°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 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.

Limitation of the Procedure

Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

Packaging

D/E Neutralizing Agar	Code No.	7375A	500 g
		7375B	2 kg
		7375C	10 kg

References

1. Engley, F. B., Jr. and B. P. Dey. 1970. A universal neutralizing medium for antimicrobial chemicals. Presented at the Chemical Specialties Manufacturing Association (CSMA) Proceedings. 56th Mid-Year Meeting.
2. Dey, B. P. and F. B. Engley, Jr. 1983. Methodology for recovery of chemically treated *Staphylococcus aureus* with neutralizing medium. Appl. Environ. Microbiol. **45**:1533-1537.
3. Dey, B. P., and F. B. Engley, Jr. 1978. Environmental sampling devices for neutralization of disinfectants, presented at the 4th International Symposium on Contamination Control.
4. Dey, B. P., and F. B. Engley, Jr. 1994. Neutralization of antimicrobial chemicals by recovery media. J. Microbiol. Methods. **19**:51-58.
5. Dey, B. P., and F. B. Engley, Jr. 1995. Comparison of Dey and Engley (D/E) Neutralizing Medium to Lethen Medium and Standard Methods Medium for recovery of *Staphylococcus aureus* from sanitized surfaces. J. Ind. Microbiol. **14**:21-25.
6. Curry, A. S., J. G. Graf, and G.N. McEwen, Jr. (eds.). 1993. CTFA Microbiology Guidelines. The Cosmetic, Toiletry and Fragrance Association, Washington, D.C.

Technical Information

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (517)372-9200 or fax us at (517)372-2006.