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D/E Neutralizing Broth
Reference 693355

Intended Use
D/E Neutralizing Broth is used to neutralize and determine the bactericidal activity of antiseptics and disinfectants. D/E Neutralizing Broth is not intended for use in the diagnosis of disease or other conditions in humans.

Description
D/E Neutralizing Broth was developed by Dey and Engley 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 Letheen media, Thioglycollate media, and Neutralizing Buffer.

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

Typical Formulation

Dextrose	10.0 g/L
Lecithin	7.0 g/L
Sodium Thiosulfate	6.0 g/L
Polysorbate 80	5.0 g/L
Enzymatic Digest of Casein	5.0 g/L
Yeast Extract	2.5 g/L
Sodium Bisulfite	2.5 g/L
Sodium Thioglycollate	1.0 g/L
Bromocresol Purple	0.02 g/L

Final pH: 7.6 ± 0.2 at 25°C
Formula is adjusted and/or supplemented as required to meet performance specifications.

Precaution
1. Refer to SDS

Preparation
1. Dissolve 39 g of the medium in one liter of purified water.
2. Mix thoroughly.
3. Autoclave at 121°C for 15 minutes.

Test Procedure
D/E Neutralizing Broth is used in a variety of procedures. Consult appropriate references for complete information.

Quality Control Specifications
Dehydrated Appearance: Powder is homogeneous, free-flowing, may form soft lumps, and bluish-gray to green.

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Prepared Appearance: Prepared medium is opaque, may appear flocculent upon sitting, and lavender to bluish-purple.
Expected Cultural Response: Cultural response in D/E Neutralizing Broth w/Tween incubated aerobically at 35 ± 2 °C and examined for growth after 18 - 48 hours.

MICROORGANISM	ATCC	APPROX. INOCULUM (CFU)	EXPECTED RESULTS
Pseudomonas aeruginosa	27853	10-100	Growth
Bacillus subtilis	9372	10-100	Growth
Escherichia coli	25922	10-100	Growth
Staphylococcus aureus	25923	10-100	Growth
Salmonella typhimurium	14028	10-100	Growth

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

Results
Refer to appropriate references and procedures for results.

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.

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. **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 Letheen 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.

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7. **Roberts D., Hooper, W. and Greenwood, M.,** (1995). Methods for the examination of food for micro-organisms of public health significance, 2nd edition, section 5.10, Practical Food Microbiology. Butler & Tanner. ISBN 0 901144 36 3.