PRODUCT SPECIFICATION SHEET

Dey-Engley Neutralizing Broth (DM792)

Intended Use
Dey-Engley Neutralizing Broth (DM792) is recommended for neutralizing and testing antiseptics and disinfectants.

Product Summary and Explanation
Dey-Engley Neutralizing Broth is formulated as per the procedure described by Engley and Dey to neutralize a broad spectrum of disinfectants and preservative antimicrobial chemicals, including quaternary ammonium compounds, phenolics, iodine, chlorine preparations, mercurials, formaldehyde, and glutaraldehyde.[1] A strongly bacteriostatic substance inhibits the growth and reproduction of bacteria without killing them. These bacteria hold the ability to cause infection under favourable conditions. D/E Neutralizing media neutralizes higher concentrations of residual antimicrobials as compared with other standard neutralizing formulations, such as Letheen media, Thioglycollate media, and Neutralizing Buffer.[2,3] Complete neutralization of disinfectants is crucial and disinfectant residues can result in a false negative (no-growth) test. Dey-Engley Neutralizing Broth is used for the neutralization and testing of antiseptics and disinfectants according to the procedure of Engley and Dey.[4] Dey-Engley Neutralizing Broth is especially suited for environmental sampling where neutralization of the chemical is important to determine its bactericidal activity. Dey-Engley Neutralizing Broth Base and Dey-Engley Neutralizing Broth have the same formula but the former does not containing the neutralizing components.

Principles of the Procedure
Dey-Engley Neutralizing Broth conatins casein enzymic hydrolysate which provide essential nutrients for metabolism. Dextrose is an energy and carbon source. Yeast extract is also a rich source of vitamin B-complex. The present formulation incorporates neutralizing substances for almost all the active products used as antiseptics and disinfectants. Sodium bisulfite neutralizes aldehydes; sodium thioglycollate neutralizes mercurials; sodium thiosulfate neutralizes iodine and chlorine,[1] lecithin neutralizes quaternary ammonium compounds; and polysorbate 80, a non-ionic surface-active agent, neutralizes substituted phenolics.[5-8] Bromocresol purple is an indicator for dextrose utilization. Due to the high concentration of lecithin in the broth medium, turbidity cannot be used to detect growth. Therefore, bromocresol purple and dextrose are added to the medium. Those organisms that ferment dextrose will turn the medium from purple to yellow. Growth of *Pseudomonas* species, which do not ferment dextrose, can be detected by the formation of a pellicle on the surface of the broth.

Formula / Liter

Ingredients	Gms / Liter
Part A	
Casein enzymic hydrolysate	5.00
Yeast extract	2.50
Dextrose	10.00
Sodium thiosulphate	6.00
Sodium thioglycollate	1.00
Sodium bisulphite	2.50
Bromocresol purple	0.02
Part B	
Polysorbate 80	5.00
Part C	
Lecithin	7.00
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 only.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

Directions

1. Dissolve 7 gm Part C separately in 50ml distilled water and boil at 100 °C for 10 minutes
2. Suspend 27.02 grams of part A + 5 grams of part B in 950 ml distilled water and add part C 50ml.
3. Heat to boiling to dissolve the medium completely.
4. Autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle. Mix well and dispense as desired.

Quality Control Specifications

Dehydrated Appearance	Light yellow to bluish grey homogeneous free flowing powder
Prepared Medium	Purple coloured, opalescent solution in tubes
Reaction of 3.9% Solution	pH : 7.6 ± 0.2 at 25°C
Gel Strength	Not Applicable

Expected Cultural Response: Cultural characteristics observed after an incubation at 35-37°C for 40-48 hours.

Sr. No.	Organisms	Results to be achieved	
		Inoculum (CFU)	Growth
1.	*Bacillus subtilis ATCC 6633*	50 -100	good-luxuriant
2.	*Escherichia coli ATCC 25922*	50 -100	good-luxuriant
3.	*Pseudomonas aeruginosa ATCC 27853*	50 -100	good-luxuriant
4.	*Salmonella Typhimurium ATCC 14028*	50 -100	good-luxuriant
5.	*Staphylococcus aureus ATCC 25923*	50 -100	good-luxuriant
6.	*Escherichia coli ATCC 8739*	50 -100	good-luxuriant
7.	*Staphylococcus aureus ATCC 6538*	50 -100	good-luxuriant

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

Test Procedure

Neutralization Test

1. Prepare two sets of test tubes, one containing 9 ml Dey-Engley Neutralizing Broth (DM792) and other with 9 ml Dey-Engley Neutralizing Broth Base (DM790), for testing disinfectants.
2. Add 1 ml of disinfectant under test. Mix well and allow it to stand for 15 minutes.
3. Inoculate 0.1 ml of 1:100,000 dilution of overnight broth cultures and incubate at 37°C for 48 hours.
4. To check bactericidal activity, both broth tubes are inoculated on D/E Neutralizing Agar (DM791).
5. Refer to appropriate references for standard test procedures.

Results

1. Growth is indicated by a colour change from purple to yellow or pellicle formation. Growth in Neutralizing Broth and no growth in Neutralizing Broth Base indicate neutralization of disinfectant.
2. Positive growth from negative tubes of Neutralizing Broth Base indicates bacteriostatic substance while negative growth indicates a bactericidal disinfectant.
3. All positive tubes should show growth on Dey-Engley Neutralizing Agar. The control disinfectants used in test procedure are 2% chlorine, 2% formaldehyde, 1% glutaraldehyde, 2% iodine, 2% phenol, 1/750 quaternary ammonium compounds, 1/1000 mercurials etc.
4. Refer to appropriate references and test procedures for interpretation of results.

Storage

Store the sealed bottle containing the dehydrated medium at 10 - 30°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 the expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedure

1. For identification, organisms must be in pure culture. Morphological, biochemical and/or serological tests should be performed for final identification.
2. Consult appropriate texts for detailed information and recommended procedures.

Packaging
Product Name : Dey-Engley Neutralizing Broth
Product Code : DM792
Available Pack sizes : 100gm/500gm

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. Engley and Dey, 1970. Chem. Spec. Manuf. Assoc. Proc., Mid-Year Meet., p. 100.
5. Downes F. P. and Ito K., (Ed.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed. American Public Health Association, Washington, D.C.
6. Quisno R.A., Gibby I.W., and Foter M.J., 1946, Am. J. Phar., 118:320.
7. Erlandson A. L., and Lawrence C. A., 1953, Science 118:274.
8. Brummer B., 1976, Appl. Environ. Microbiol., 32:80.

Further Information

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