D/E NEUTRALIZING BROTH

Cat. no. K108	D/E Neutralizing Broth, 16x125mm Tube, 10ml	20 tubes/box
Cat. no. U75	D/E Neutralizing Broth, 180ml Wide Mouth Jar, 90ml	12 jars/box
Cat. no. U76	D/E Neutralizing Broth, 1L Polycarbonate Bottle, 750ml	10 bottles/box
Cat. no. U332	D/E Neutralizing Broth, 60ml Polypropylene Bottle, 45ml	25 bottles/box

INTENDED USE

Hardy Diagnostics D/E Neutralizing Broth is utilized to neutralize antiseptics and disinfectants as well as to detect organisms remaining after treatment. This broth is especially suited for environmental sampling where neutralization of the chemical is important to determining the bactericidal activity.

This product is not intended to be used for the diagnosis of human disease.

SUMMARY

D/E Neutralizing Broth, also known as Dey-Engley Neutralizing Broth, is capable of neutralizing a broad spectrum of antiseptic and disinfectant chemicals including quaternary ammonium compounds, phenolics, iodine, chlorine preparations, mercurials, formaldehyde and glutaraldehyde. It can determine the bactericidal capability of disinfectants and therefore is well suited for environmental sampling.

D/E Neutralizing Broth contains various neutralizing agents: lecithin, Tween®, sodium thiosulfate, and sodium bisulfite. Lecithin neutralizes quaternary ammonia compounds while phenolic disinfectants and hexachlorophene are neutralized by Tween®. Together, lecithin and Tween® neutralize ethanol. Sodium thiosulfate neutralizes iodine and chlorine, where as sodium bisulfite neutralizes formaldehyde and gluteraldehyde.

Complete neutralization of disinfectants is vital as disinfectant carryover can cause a false no-growth test result. D/E Neutralizing media effectively neutralizes the inhibitory effects of disinfectant carryover, allowing differentiation between bacteriostasis and the true bactericidal action of disinfectants.

In addition to neutralizing agents, this media contains ingredients that enhance the growth of a wide variety of microorganisms. Pancreatic digest of casein provides the carbon and nitrogen sources. Yeast extract provides vitamins and growth factors required for growth. Dextrose is added as a fermentable carbohydrate source. Bromcresol purple is added to the media as a colormetric indicator to demonstrate the production of acid from dextrose.

FORMULA

Ingredients per liter of deionized water:*

Dextrose	10.0gm

Lecithin	7.0gm
Sodium Thiosulfate	6.0gm
Pancreatic Digest of Casein	5.0gm
Tween® 80	5.0gm
Yeast Extract	2.5gm
Sodium Bisulfite	2.5gm
Sodium Thioglycollate	1.0gm
Monopotassium Phosphate	0.1gm
Bromcresol Purple	0.02gm

Final pH 7.6 +/- 0.3 at 25°C.

* Adjusted and/or supplemented as required to meet performance criteria.

STORAGE AND SHELF LIFE

Storage: Upon receipt store at 2-8ºC. away from direct light. Media should not be used if there are any signs of deterioration, discoloration, contamination, or if the expiration date has passed. Product is light and temperature sensitive; protect from light, excessive heat and freezing.

The expiration dating on the product label applies to the product in its intact packaging when stored as directed. The product may be used and tested up to the expiration date on the product label and incubated for the recommended quality control incubation times.

Refer to the document "Storage" for more information.

PRECAUTIONS

This product may contain components of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not guarantee the absence of transmissible pathogenic agents. Therefore, it is recommended that these products be treated as potentially infectious, and handle observing the usual universal blood precautions. Do not ingest, inhale, or allow to come into contact with skin.

This product is for laboratory use only. It is to be used only by adequately trained and qualified laboratory personnel. Observe approved biohazard precautions and aseptic techniques. All laboratory specimens should be considered infectious and handled according to "standard precautions." The "Guidelines for Isolation Precautions" is available from the Centers for Disease Control and Prevention at www.cdc.gov/ncidod/dhqp/gl_isolation.html.

For additional information regarding specific precautions for the prevention of the transmission of all infectious agents from laboratory instruments and materials, and for recommendations for the management of exposure to infectious disease, refer to CLSI document M-29: *Protection of Laboratory Workers from Occupationally Acquired Infections: Approved Guideline.*

Sterilize all biohazard waste before disposal.

Refer to the document "Precautions When Using Media" for more information.

Refer to the document SDS Search instructions on the Hardy Diagnostics' website for more information.

PROCEDURE

Specimen Collection: Consult listed references for appropriate methods for the collection of specimens from environmental and industrial sources.

Method of Use: Refer to listed references of test protocols and additional procedures using this media. Inoculate the media as desired and incubate with caps loosened, under aerobic conditions at 35ºC. for 24-48 hours.

INTERPRETATION OF RESULTS

Colonies of dextrose fermenting microorganisms will cause the media to turn yellow in color.

Consult listed references for information pertaining to colony morphology and biochemical tests required for identification.[1-5]

LIMITATIONS

It is recommended that biochemical, immunological, molecular, or mass spectrometry testing be performed on colonies from pure culture for complete identification.

Refer to the document "Limitations of Procedures and Warranty" for more information.

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as loops, other culture media, swabs, applicator sticks, incinerators, and incubators, etc., as well as serological and biochemical reagents, are not provided.

QUALITY CONTROL

Hardy Diagnostics tests each lot of commercially manufactured media using appropriate quality control microorganisms and quality specifications as outlined on the Certificates of Analysis (CofA). The following organisms are routinely used for testing at Hardy Diagnostics:

Test Organisms	Inoculation Method*	Incubation			Results
		Time	Temperature	Atmosphere	
Bacillus subtilis ATCC® 6633	A	40-48hr	35°C	Aerobic	Growth; usually no color change
Escherichia coli ATCC® 25922	A	40-48hr	35°C	Aerobic	Growth; yellow color change
Pseudomonas aeruginosa ATCC® 9027	A	40-48hr	35°C	Aerobic	Growth; no color change
Salmonella enterica ATCC® 14028	A	40-48hr	35°C	Aerobic	Growth; yellow color change
Staphylococcus aureus ATCC® 6538	A	40-48hr	35°C	Aerobic	Growth; yellow color change
Additional test organism for Cat. no. U332					
Streptococcus mutans ATCC® 35668	A	40-48hr	35°C	Aerobic	Growth; yellow color change

* Refer to the document "Inoculation Procedures for Media QC" for more information.

USER QUALITY CONTROL

End users of commercially prepared culture media should perform QC testing in accordance with applicable government regulatory agencies, and in compliance with accreditation requirements. Hardy Diagnostics recommends end users check for signs of contamination and deterioration and, if dictated by laboratory quality control procedures or regulation, perform quality control testing to demonstrate growth or a positive reaction and to demonstrate inhibition or a negative reaction, if applicable. Hardy Diagnostics quality control testing is documented on the certificates of analysis (CofA) available from Hardy Diagnostics Certificates of Analysis website. In addition, refer to the following document "Finished Product Quality Control Procedures," for more information on QC or see reference(s) for more specific information.

PHYSICAL APPEARANCE

D/E Neutralizing Broth should appear opaque, with an even suspension of particulates, and lavender to purple in color.

Bacillus subtilis (ATCC® 6633) growing in D/E Neutralizing Broth (Cat. no. K108). Incubated aerobically for 24 hours at 35ºC. No color change to yellow was indicative as negative for acid production from dextrose.

Escherichia coli (ATCC® 25922) growing in D/E Neutralizing Broth (Cat. no. K108). Incubated aerobically for 24 hours at 35ºC. The yellow color change was indicative as positive for acid production from dextrose.

REFERENCES

1. U.S. Food and Drug Administration. *Bacteriological Analytical Manual.* AOAC, Arlington, VA. http://www.fda.gov/Food/FoodScienceResearch/LaboratoryMethods/ucm2006949.htm.

2. American Public Health Association. *Standard Methods for the Examination of Dairy Products,* APHA, Washington, D.C.

3. APHA Technical Committee on Microbiological Methods for Foods. *Compendium of Methods for the Microbiological Examination of Foods,* APHA, Washington, D.C.

4. American Public Health Association. *Standard Methods for the Examination of Water and Wastewater,* APHA, Washington, D.C.

5. *United States Pharmacopoeia and National Formulary* (USP-NF). Rockville, MD: United States Pharmacopeial Convention.

ATCC is a registered trademark of the American Type Culture Collection.
Tween is a registered trademark of ICI Americas, Inc.

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The Hardy Diagnostics manufacturing facility and quality management system is certified to ISO 13485.

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