CRITERION™ NEUTRALIZING BUFFERED PEPTONE WATER

Cat. no. C9240	CRITERION™ Neutralizing Buffered Peptone Water (nBPW)	56g
Cat. no. C9241	CRITERION™ Neutralizing Buffered Peptone Water (nBPW)	500g
Cat. no. C9242	CRITERION™ Neutralizing Buffered Peptone Water (nBPW)	2kg
Cat. no. C9243	CRITERION™ Neutralizing Buffered Peptone Water (nBPW)	10kg
Cat. no. 00084	Sodium Bicarbonate**	1kg
** sold separately		

INTENDED USE

Hardy Diagnostics CRITERION™ Neutralizing Buffered Peptone Water (nBPW) is recommended for use in the recovery of sub-lethally injured *Salmonella* species from industrial samples prior to selective enrichment and isolation.

Dehydrated culture media is a raw material not intended for use in the diagnosis of human disease. For implementation, this product requires additional processing and supplementation of ingredients before use.

SUMMARY

Salmonella spp. may be present in foods, particularly poultry products, yet cells may be sub-lethally injured by food processing techniques. Consequently, it may be difficult to recover injured cells of this organism using selective media and the organism may go undetected using traditional culture techniques. Beginning July 1, 2016, the United States Department of Agriculture, Food Safety and Inspection Service (USDA FSIS) instituted new guidelines for the development of Neutralizing Buffered Peptone Water (nBPW) to aid in the recovery of *Salmonella* spp. from domestic and imported poultry verification sampling, including chicken carcass rinses, poultry parts rinses and young turkey carcass sponge swabs.[6] The USDA FSIS also states nBPW is safe as a direct rinse or swab, and should be used as a non-selective pre-enrichment medium to promote the recovery of sub-lethally injured bacteria, particularly *Salmonella* spp.[6]

CRITERION™ nBPW contains peptones that act as nitrogenous compounds to promote bacterial growth. Phosphates in the buffer help to maintain pH when combined with sodium bicarbonate during preparation. Maintenance of pH is important when attempting to recover sub-lethally injured cells, because a low pH can be detrimental to the repair and growth of damaged microorganisms. In addition, CRITERION™ nBPW contains neutralizing agents to reduce the inhibitory effects of carryover from antimicrobial interventions.[6] The medium requires the addition of sodium bicarbonate prior to use to conform to the USDA FSIS nBPW formulation.

FORMULA*

Gram weight per liter:	28.0g/L
Buffered Peptone Water (BPW)	20.0g
Lecithin	7.0g
Sodium Thiosulfate	1.0g

Final pH 7.7 +/- 0.5 at 25ºC.

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

STORAGE AND SHELF LIFE

Store the sealed bottle(s) that contain dehydrated culture medium at 2-30ºC. Dehydrated culture medium is very hygroscopic and will clump when exposed to moisture and air. Keep lid tightly sealed. Protect dehydrated culture media from moisture and light. Dehydrated culture media should be discarded if clumped, if the media is not free-flowing or if the color has changed from its original beige

Store the prepared culture media at 2-30ºC and do not remove the container desiccant, if applicable.

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.

METHOD OF PREPARATION FOR DEHYDRATED CULTURE MEDIA

1. Suspend 28g of the dehydrated culture media in 833mL of distilled or deionized water. Stir to mix thoroughly to dissolve.

3. Sterilize in the autoclave at 121ºC. for 15 minutes.

4. Cool to less than 55ºC.

5. To adjust pH, dissolve 12.5g of sodium bicarbonate (Cat. no. 00084) into 167mL of purified water and filter sterilize.

6. Add the filter sterlized sodium bicarbonate solution to the 833mL of pre-sterilized broth.

7. Dispense as desired into pre-sterilized containers.

Note: The shelf life of in-house prepared media from dehydrated culture media is dependent upon preparation methods, container quality, equipment, storage conditions, and batch testing criteria and must be validated by the end user. Refer to *USP Microbiological Best Laboratory Practices <1117>* for more information on validation procedures.[1]

PROCEDURE

Sample Collection: Consult reference methods for complete procedures on sample collection.[1-6]

Method of Use: Gently mix nBPW prior to use. Consult listed references for complete procedures for handling poultry carcasses prior to rinsing and swabbing and for information on the recovery of *Salmonella* spp. from food or poultry samples.[1,2,5,6]

1. Inoculate 10g or 10mL of sample for every 50mL of Neutralizing Buffered Peptone Water (nBPW).

2. Incubate at 35ºC. for 18 to 24 hours.

3. Transfer 10mL of the incubated sample to 100mL of Tetrathionate Broth (Cat. no. U165) and incubate at 35ºC. Other selective enrichments may be used.[1,2,5,6]

4. After 24 and 48 hours, subculture to Brilliant Green Agar (Cat. no. G75), XLD Agar (Cat. no. G65) and/or HE Agar (Cat. no. G63) and incubate plates for 18 to 24 hours at 35ºC. NOTE: It is recommended that more than one selective agar be used in parallel to ensure recovery when salmonellae are present, since no single medium is appropriate in all situations.[1,2,5,6]

5. Examine plates for typical colonies of *Salmonella* spp. and perform further testing for complete identification.

INTERPRETATION OF RESULTS

Consult listed references for appropriate interpretation of results.[1-6]

Following incubation, examine solid media for growth and typical colony morphology.

LIMITATIONS

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

Some formulations may require a settling period before pH testing of the prepared medium. If the pH is tested immediately after preparation and is out of specification, retest the medium after 24 hours to obtain final pH results.

nBPW is a non-selective medium. Overgrowth of competing flora in the test sample may affect recovery of salmonellae.

To recover *Campylobacter* spp. from prepared batches of CRITERION™ nBPW, subculture samples to an appropriate agar medium, such as Chocolate Agar, and incubate plates under microaerophilic conditions.

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, sodium bicarbonte (Cat. no. 00084), other culture media such as Brilliant Green Agar (Cat. no. G75), XLD Agar (Cat. no. G65), HE Agar (Cat. no. G63), Chocolate Agar (Cat. no. E14), or Tetrathionate Broth (Cat. no. U165), 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	
Salmonella enterica ATCC® 14028	A	18-24hr	35°C	Aerobic	Growth and typical colony morphology upon subculture to XLD Agar
Campylobacter jejuni ATCC® 33291	0.5 MF	18-24hr	35°C	Tight Cap	Growth when subcultured to Chocolate Agar and incubated under microaerophilic conditions
Escherichia coli ATCC® 25922	A	18-24hr	35°C	Aerobic	Partial to complete inhibition upon subculture to XLD Agar

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

USER QUALITY CONTROL

Users of dehydrated 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

CRITERION™ Neutralizing Buffered Peptone Water (nBPW) powder should appear homogeneous, free-flowing, and beige in color. The prepared medium should appear opaque, cloudy, and light yellow in color.

REFERENCES

1. Juven, B.J., N. Cox, J.S. Bailey, J.E. Thomson, O.W. Charles, and J.V. Schutze. 1984. Recovery of *Salmonella* from artificially contaminated poultry feeds in non-selective and selective broth media. *Jour. of Food Prot.* ; 47:299-302.

2. Sadovski, A.Y. 1977. *J. Food Technology*; 12:85-91.

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

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

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

6. USDA FSIS. June 8, 2016. FSIS Notice 41-16. Washington D.C

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

HDQA 2207B [C]