Buffered Peptone Water ISO

Intended Use
Buffered Peptone Water ISO is recommended as a pre-enrichment medium of *Enterobacteriaceae* organisms such as *Salmonella* and *Cronobacterium* species from food and animal feeding stuffs, water, milk, milk products and other products in compliance with ISO specifications ISO 6579-1:2017, ISO 6887-1:2017, ISO 21528-1:2017, ISO 22964:2017.

Summary
Microorganisms that are subjected to environmental stresses may become structurally or metabolically damaged or injured. These microorganisms are unable to replicate in selective environments. Therefore, these injured organisms must be resuscitated or permitted to repair the damage by incubation in an appropriate, non-selective environment. This medium is also recommended by APHA for pre-enrichment of *Salmonella*, *Cronobacter* and *Listeria*. Edel and Kampelmacher noted that sub-lethal injury to Salmonellae may occur in many food preservation processes. Pre-enrichment in Buffered Peptone Water at 35°C for 18-24 hours results in repair of injured cells. The buffering system prevents bacterial damage due to change in the pH of the medium. ISO committee has also recommended this pre-enrichment medium for the detection of *Enterobacteriaceae*, *Salmonella, Cronobacter* and *Listeria* species from food stuffs and other materials. It is also recommended as a diluent for enumerations of all microorganisms.

Principle
Enzymatic Digest of Casein (Peptone) serves as a source of carbon, nitrogen, vitamins and minerals. Sodium chloride provides sodium ions for the membrane transport and maintains osmotic equilibrium of the medium. Phosphates buffers the medium.

Formula*

Ingredients	**g/L**
Enzymatic Digest of Casein (Peptone)	10.0
Sodium Chloride	5.0
Disodium Hydrogen Phosphate, Dodecahydrate ($Na_2HPO_4.12H_2O$)	9.0**
Potassium Dihydrogen Phosphate	1.5
Final pH (at 25°C)	7.0 ± 0.2

*Adjusted to suit performance parameters.
**9.0 g of $Na_2HPO_4.12H_2O$ is equivalent to 3.57 g of Na_2HPO_4 Anhydrous.

Storage and Stability
Store dehydrated medium below 30°C in tightly closed container and the prepared medium at 2ºC-8°C. Avoid freezing and overheating. Use before expiry date on the label. Once opened keep powdered medium closed to avoid hydration.

Type of specimen
Food and dairy samples.

Specimen Collection and Handling
Ensure that all samples are properly labelled.
Follow appropriate techniques for handling samples as per established guidelines.
Some samples may require special handling, such as immediate refrigeration or protection from light, follow the standard procedure.
The samples must be stored and tested within the permissible time duration.
After use, contaminated materials must be sterilized by autoclaving before discarding.

Directions

1. Suspend 20.07 g (Equivalent weight of dehydrated medium) of the powder in 1000 mL purified / distilled water.
2. Mix thoroughly.
3. Warm slightly with frequent agitation to dissolve the powder completely and dispense in 50 mL amounts.
4. Sterilize by autoclaving at 121°C (15 psi) for 20 minutes as per validated cycle.

Quality Control

Dehydrated Appearance: Cream to Light yellow coloured, homogenous, free flowing powder.
Prepared Appearance: Light yellow coloured, clear solution without any precipitate.
Cultural Response: Cultural response is observed after an incubation at 37°C ± 1°C for 18 ± 2 hours.

Pre-enrichment for *Salmonella* and *Enterobacteriaceae* detection

Organisms (ATCC)	Inoculum cfu	Expected Growth
Salmonella enterica subsp. *enterica* serovar *Typhimurium* (14028)	$>10^4$	Good
Escherichia coli (25922)	$>10^4$	Good
Escherichia coli (8739)	$>10^4$	Good

Diluent for enumeration of microorganisms

Organisms (ATCC)	Inoculum cfu	Expected Growth
Escherichia coli (25922)	$>10^4$	T1 plate counts w/in ± 30% of counts for T0
Escherichia coli (8739)	$>10^4$	T1 plate counts w/in ± 30% of counts for T0
Listeria monocytogenes strain Li 23 (19114)	$>10^4$	T1 plate counts w/in ± 30% of counts for T0
Listeria monocytogenes serotype 4b (19115)	$>10^4$	T1 plate counts w/in ± 30% of counts for T0

Note: For strains tested as diluent, a satisfactory result is represented by recovery of 30% of the control cfu (T0) (0 minutes) from an inoculum of 50-150 colony forming units (cfu) after holding at 20-25°C for 45 minutes (T1) for *Escherichia coli* and from an inoculum of 50-100 cfu after holding at 18-22°C for 1 hour (T1) for *Listeria monocytogenes*.
For use as non-selective pre-enrichment broth, a satisfactory result is represented by visible growth from an inoculum of 10-100 cfu. All ISO/CEN 11133-2 control strains are included in the test panel.

Interpretation of Results

Growth in the medium is indicated by the presence of turbidity compared to an uninoculated control.

Performance and Evaluation

Performance of the product is dependent on following parameters as per product label claim:

1. Directions
2. Storage
3. Expiry

Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

Reference

1. Edel and Kampelmacher, 1973, Bull. W.H.O., 48:167.
2. Sadovski, 1977, J. Food Technol., 12:85.
3. Juven, Cox, Bailey, Thomson, Charles and Schutze, 1984, J. Food Prot., 47:299
4. Microbiology of the food chain- Preparation of test samples, initial suspension and decimal dilutions for microbiological examination - Part 1 General rules for the preparation of the initial suspension and decimal dilutions. International Organization for Standardization (ISO), 6887-1:2017.
5. Microbiology of the food chain - Horizontal method for the detection and enumeration of Enterobacteriaceae - Part 1: Detection of Enterobacteriaceae. International Organization for Standardization (ISO), ISO 21528-1:2017.

6. Microbiology of the food chain - Horizontal method for the detection, enumeration and serotyping of Salmonella - Part I Detection of Salmonella. International Organization for Standardization (ISO), ISO/DIS 6579-1:2017.
7. Microbiology of the food chain - Horizontal method for the detection of Cronobacter spp. International Organization for Standardization. Draft ISO/ TS 22964, 2017.
8. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat No.	Product description	Pack Size
201020390100	Dehydrated Culture Media (ISO)	100 g
201020390500	Dehydrated Culture Media (ISO)	500 g
201020392500	Dehydrated Culture Media (ISO)	2.5 k