

Modified Buffered Peptone Water with Pyruvate (NCM0084)

Intended Use

Modified Buffered Peptone Water with Pyruvate (mBPWp) is used for the isolation of Enterohemorrhagic *E. coli* (EHEC) in a laboratory setting. Modified Buffered Peptone Water with Pyruvate (mBPWp) is not intended for use in the diagnosis of disease or other conditions in humans.

Description

Enterohemorrhagic *E. coli* (EHEC) are recognized as the primary cause of hemorrhagic colitis (HC) or bloody diarrhea. This infection can also lead to hemolytic uremic syndrome (HUS). Enterohemorrhagic infections are usually food or water borne and have been implicated in undercooked ground beef, raw milk, lunchmeat, water, unpasteurized apple cider, sprouts and vegetables.

Modified Buffered Peptone Water with Pyruvate is recommended in the sample preparation for the screening method following FDA/BAM Enterohemorrhagic *E. coli* (EHEC) Protocol. After the sample is incubated for five hours in Modified Buffered Peptone Water with Pyruvate, Cefsulodin, Acriflavin and Vancomycin are added, followed by an additional incubation period. These antimicrobial supplements effectively suppress normal flora, while allowing the growth of viable O157:H7 cells (including STEC) and is capable of detecting <1 cfu/g in foods. Improved recovery of pathogenic *E. coli* from difficult commodities was achieved with Modified Buffered Peptone Water with Pyruvate enrichment.

Typical Formulation

Enzymatic Digest of Casein	10.0 g/L
Lactose	10.0 g/L
Yeast Extract	6.0 g/L
Acid Digest of Casein	5.0 g/L
Sodium Chloride	5.0 g/L
Sodium Phosphate, dibasic	3.6 g/L
Potassium Phosphate, monobasic	1.5 g/L
Sodium Pyruvate	1.0 g/L

Final pH: 7.2 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precaution

Refer to SDS

Preparation

1. Dissolve 42.1 g of the medium in one liter of purified water.
2. Mix thoroughly.
3. Autoclave at 121°C for 15 minutes.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, free flowing, and light beige.

Prepared Appearance: Prepared medium is clear and light to dark amber.

Expected Cultural Response: Cultural response in Modified Buffered Peptone Water with Pyruvate incubated aerobically at $37 \pm 1^\circ\text{C}$ and examined for growth at 18 - 24 hours.

Microorganism	Approx. Inoculum (CFU)	Expected Results
<i>Escherichia coli</i> ATCC® 25922	10 - 300	Good growth
<i>Escherichia coli</i> O157:H7 ATCC® 35150	10 - 300	Good growth
<i>Salmonella typhimurium</i> ATCC® 14028	10 - 300	Good growth

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

Test Procedure

Refer to the BAM Manual for complete directions on sample preparation. After the sample has been prepared, material will be placed in Modified Buffered Peptone Water with Pyruvate and incubated at $37 \pm 1^\circ\text{C}$ for 5 hours. Next add 1 mL each of Acriflavin (10 mg/L), Cefsulodin (10 mg/L), Vancomycin (8 mg/L) and incubate homogenate at $42 \pm 1^\circ\text{C}$ for 18 – 24 hours.

Results

Refer to FDA, Bacteriological Analytical Manual (BAM).

Expiration

Refer to 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.

Limitation of the Procedure

Due to nutritional variation, some strains may be encountered that grow poorly.

Storage

Store sealed bottle containing the dehydrated medium at 2 - 30°C. 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. www.fda.gov/Food/ScienceResearch/LaboratoryMethods/BacteriologicalAnalyticalManualBAM/default.htm.
2. Smart Cycler Operator Manual. 1999. USA.
3. Weagant, S. D. and A. J. Bound. 2001. Comparison of Methods for Enrichment and Isolation of *Escherichia coli* O157:H7 from Artificially Contaminated Salad Mixes. Laboratory Information Bulletin, LIB 4258, Aug. 2001.