Buffered Peptone Water EP/USP

Cat. 1401

Recommended as a diluent for the homogenization of samples.

Practical information

Aplications	Categories
Diluent	General use

Industry: Pharmaceutical/Veterinary / Final product Quality Control

Regulations: USP / European Pharmacopoeia

Principles and uses

Buffered Peptone Water is recommended by the European Pharmacopoeia in the paragraph 2.6.12 "Microbiological examination of non – sterile products: Microbial enumeration test', and in the paragraph 2.6.13 "Microbiological examination of non-Sterile products: test for specified micro-organisms" as a diluent for the homogenization of samples in the examination of TAMC and TYMC, and specified microorganisms in products.

Pancreatic digest of casein provides nitrogen, vitamins, minerals and amino acids essential for growth. Potassium phosphates act as a buffer system and Sodium chloride supplies essential electrolytes for transport and osmotic balance.

It is used in the preparation of the samples to dissolve or dilute water-soluble products (1:10 dilution) and non-fatty products insoluble in water (in general 1:10 dilution, but some may require larger volumes of Buffered Peptone Water) Fatty products are homogenized with a suitable sterile surface-active agent such as polysorbate or tween 80 heated if necessary to no more than 40ºC or, in exceptional cases, to no more than 45ºC. They are mixed carefully and if necessary the temperature is maintained in a water-bath. It is added enough of the pre-warmed Buffered Peptone Water diluent to make a 1:10 dilution of the original product.

Formula in g/L

Pancreatic digest of casein	1	Potassium dihydrogen phosphate	3,6
Sodium chloride	4,3	Disodium hydrogen phosphate dihydrate	7,2

Preparation

Suspend 16,1 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Dispense into appropriate containers and sterilize in autoclave at 121 °C for 15 minutes.

Instructions for use

According to European Pharmacopoeia for the examination of TAMC and TYMC in products :

Membrane filtration:
- Prepare the product sample suspending, dissolving or diluting the product to be examined in the Buffered Peptone Water.
- Transfer the appropriate amount of the sample to a membrane filter.
- Place the membrane to the surface of Trypticasein Soy Agar (Cat. 1068) in case of TAMC or Sabouraud Dextrose Agar (Cat. 1024) in case of TYMC.
- Incubate the plate of Trypticasein Soy Agar (Cat. 1068) at 30-35 ºC for 3-5 days and the plate of Sabouraud Dextrose Agar (Cat. 1024) at 20-25 ºC for 5-7 days.

Plate-count methods:
- Prepare the product sample suspending, dissolving or diluting the product to be examined in the Buffered Peptone Water.
- Inoculate the plates of Trypticasein Soy Agar (Cat. 1068) in case of TAMC or Sabouraud Dextrose Agar (Cat. 1024) in case of TYMC, conforming to the

pour-plate method or the surface-spread method.
- Incubate the plates of Trypticasein Soy Agar (Cat. 1068) at 30-35 ºC for 3-5 days and the plates of Sabouraud Dextrose Agar (Cat. 1024) at 20-25 ºC for 5-7 days.
- Select the plates corresponding to a given dilution and showing the highest number of colonies less than 250 (TAMC) or 50 (TYMC).

Most-probable number method (only for TAMC):
- Prepare and dilute the product sample to be examined in Buffered Peptone Water, and inoculate into tubes of Trypticasein Soy Broth (Cat 1224).
- Incubate all tubes at 30-35 ºC for 3-5 days.
- Record for each level of dilution the number of tubes that showing growth and determinate the most probable number of microorganisms.

Quality control

Solubility	Appareance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25ºC)
w/o rests	Fine powder	Whitish	Transparent	7,0±0,2

Microbiological test

Incubation conditions: (30-35ºC /18-24 h).

Microorganisms	Specification
Candida albicans ATCC 10231	Good growth
Salmonella typhimurium ATCC 14028	Good growth
Aspergillus brasiliensis ATCC 16404	Good growth
Staphylococcus aureus ATCC 6538	Good growth
Bacillus subtilis ATCC 6633	Good growth
Escherichia coli ATCC 8739	Good growth

Storage

Temp. Min.:2 ºC
Temp. Max.:25 ºC

Bibliography

European Pharmacopoeia 9.0