

BUFFERED PEPTONE WATER

AGPT-OEP-500

- **Principle**

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 40 °C or, in exceptional cases, to no more than 45 °C. 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.

- **Regulatory compliance**

This product is manufactured under a quality management system in accordance with ISO 9001 and ISO 13485, and its formulation and quality control comply with applicable international standards, such as ISO 11133, where relevant.

For this specific medium, compliance is also established with the relevant requirements of USP as well as the European Pharmacopoeia.

- **Composition**

Ingredients	g/L
Pancreatic digest of casein	1.00
Potassium dihydrogen phosphate	3.60
Sodium chloride	4.30
Di-sodium hydrogen phosphate dihydrate	7.20

- **Preparation**

Suspend 16,1 grams of the medium in one litre 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.

- **Applications and 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 (AGST-0EP-500) in case of TAMC or Sabouraud Dextrose Agar (AGDS-0EP-500) in case of TYMC.
- Incubate the plate of Trypticasein Soy Agar (AGST-0EP-500) at 30-35 °C for 3-5 days and the plate of Sabouraud Dextrose Agar (AGDS-0EP-500) at 20-25 °C 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 (AGST-0EP-500) in case of TAMC or Sabouraud Dextrose Agar (AGDS-0EP-500) in case of TYMC, conforming to the pour-plate method or the surface-spread method.
- Incubate the plates of Trypticasein Soy Agar (AGST-0EP-500) at 30-35 °C for 3-5 days and the plates of Sabouraud Dextrose Agar (AGDS-0EP-500) at 20-25 °C 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 (STRB-0EP-500).
- Incubate all tubes at 30-35 °C 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	w/o rests
Appearance	Fine powder
Colour of the dehydrated medium	Whitish
Colour of the prepared medium	Transparent
Final pH (25 °C)	7.0 ± 0.2

- **Microbiological test**

Incubation conditions: 30-35 °C /18-24 h.

Microorganisms	ATCC	Specification
<i>Staphylococcus aureus</i>	6538	±30% of original count for the dilution test
<i>Salmonella typhimurium</i>	14028	±30% of original count for the dilution test
<i>Escherichia coli</i>	8739	±30% of original count for the dilution test

- **Storage**

The product is highly hygroscopic; keep the container always closed and store it properly as per the conditions mentioned on the label. The declared expiry is valid only when stored as per the conditions mentioned on the label. Temp. Min.:2 °C Temp. Max.:25 °C.

Note: Sterilize media immediately after reconstitution.

- **Bibliography**

European Pharmacopoeia 9.0

- **Product use limitation**

This product is developed, designed and supplied exclusively for research use only. It is not intended for diagnostic applications or drug development, and it is not suitable for administration to humans or animals.