

# **Product Information**

# Trypsin **10X** Solution

2.5% trypsin, gamma irradiated CATALOG NO. 59427C

## Description

Trypsin is a porcine pancreas-derived enzyme that is commonly used for the dissociation and disaggregation of anchorage-dependent mammalian cells and tissues. The concentration of trypsin necessary to dislodge cells from their substrate is dependent primarily on the cell type and the age of the culture.

This 10X concentrated solution can be diluted with calciumand magnesium-free Dulbecco's Phosphate Buffered Saline (DPBS).

SAFC Biosciences has validated a process using gamma radiation to significantly reduce the risks associated with adventitious agents such as Porcine Parvovirus (PPV), Porcine Respiratory and Reproductive Syndrome (PRRS) and Mycoplasma hyorhinis while maintaining product performance. The use of gamma irradiated trypsin requires no change to the end user's methods or procedures while giving additional assurance against microbial contaminants associated with animal-derived products.

All trypsin is obtained from the United States or other countries deemed free Bovine Spongiform Encephalopathy (BSE).

#### **Precautions**

Use aseptic technique when handling or supplementing this solution. This product is for further manufacturing use. THIS PRODUCT IS NOT INTENDED FOR HUMAN OR THERAPEUTIC USE.

## Storage

Store trypsin at -10 to -40 C. Do not use after expiration date. Repeated cycles of freezing and thawing reduce enzymatic activity and should be avoided.

#### **Formulation**

Component (all components measured in mg/L)	
INORGANIC SALTS	
Potassium chloride	400.000
Potassium phosphate monobasic anhydrous	60.000
Sodium chloride	8000.000
Sodium phosphate dibasic anhydrous	47.680
OTHER	
Dextrose anhydrous	1000.000
Phenol red sodium salt	10.620
Trypsin porcine (1:250)	25000.000

#### Indications of Deterioration

Trypsin solutions should be clear of particulates and flocculent material. Do not use if solution is cloudy or contains precipitate. Other evidence of deterioration may include color change or degradation of physical or performance characteristics.

## **Preparation Instructions**

#### **Dilution Instructions for 10X Solution**

- 1. Frozen products can either be thawed in a 37 C water bath or overnight at 2 to 8 C.
- 2. Aseptically transfer 100 mL of 10X trypsin to a sterile 1000 mL container.
- 3. Add 800 mL of a sterile calcium and magnesium-free 1X balanced salt solution (as listed below) to the container.

Dulbecco's Phosphate Buffered Saline (DPBS) (Catalog No. 59321C), or Hanks' Balanced Salt Solution (HBSS) (Catalog No. 55025C).

- 4. Mix well for several minutes.
- 5. Determine the pH of a small sample. If necessary, adjust the pH to 7.2 7.8 with 1N HCl or 1N NaOH.
- 6. Bring the final volume up to 1000 mL with the sterile salt solution and dispense into smaller volumes.
- 7. Store trypsin at -10 to -40 C.

## Methods for Use

- 1. Frozen products can either be thawed in a 37 C water bath or overnight at 2 to 8 C.
- 2. Aspirate the spent medium from the culture vessel and discard.
- 3. Rinse the monolayer with either a small amount of trypsin or a calcium and magnesium-free balanced salt solution, aspirate and discard.
- 4. Add enough trypsin solution, prewarmed in a 37 C water bath, to completely cover the cell monolayer.
- 5. Incubate the flask at 37 C, or for more sensitive cultures, at room temperature or 2 to 8 C.
- 6. When the trypsinization process is complete, cells will appear rounded upon microscopic examination and the solution in the flask will appear cloudy. Check the flask often to avoid overexposure which can damage the cells.
- 7. The trypsin should be neutralized either with serum containing medium or trypsin inhibitor. Gently centrifuge the cell suspension and discard the trypsin-containing supernatant.
- 8. Resuspend the cell pellet with fresh medium and count or culture as desired.

## Characteristics

## Adventitious Viral Agents (AVA) (PPV)

None detected in bulk powder raw material

### **Appearance**

Clear orange-red solution

**Endotoxin (at 1X concentration)** 

≤ 100.0 EU/mL

### **Enzymatic Activity**

Cells dislodged ≤ 30 minutes

#### Mycoplasma

None detected

## Osmolality (at 1X concentration)

270 - 325 mOsm/kg H<sub>2</sub>O

pH (at 25 C)

7.0 - 7.7

#### Sterility

No microbial growth detected

## References

- 1. Hodges, G. M., Linvingston, D. C., and Franks, L. M., *J. Cell Sci.* (1973) 12:887.
- 2. McKeehan, W. L., Cell Biol. Intl. Rep. (1977) 1:335.
- 3. Safton, B. M. and Rubin, H., Nature (1970) 227:843.

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