

# Product Information

## Trypsin — EDTA 1X Solution

0.05% trypsin, 0.02% EDTA, gamma irradiated

CATALOG NO. 59417C

### 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.

Ethylenediaminetetracetic acid (EDTA), a chelating agent, is added to trypsin solutions to enhance enzymatic activity by neutralizing calcium and magnesium ions that enhance cell-to-cell adhesion and obscure the peptide bonds on which trypsin acts.

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 of 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)	
<b>INORGANIC SALTS</b>	
EDTA 4Na	200.000
Potassium chloride	400.000
Potassium phosphate monobasic anhydrous	60.000
Sodium bicarbonate	350.000
Sodium chloride	8000.000
Sodium phosphate dibasic anhydrous	47.680
<b>OTHER</b>	
Dextrose anhydrous	1000.000
Phenol red sodium salt	10.620
Trypsin porcine (1:250)	500.000

### 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.

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## 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 red solution

### Endotoxin

≤ 100.0 EU/mL

### Enzymatic Activity

Cells dislodged ≤ 30 minutes

### Mycoplasma

None detected

### Osmolality

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

### pH (at 25 C)

7.2 - 8.0

### 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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