

## UAB "LT Biotech"

Į.K. 302303586, PVM kodas LT 100004741118,  
Rugių 21-24, LT-08419, Vilnius Reg. Nr. 127918, V.Į.  
Registrų centras Vilniaus filialas  
Tel/fax +370 5 216 02 27

### Trypsin-EDTA (0.25 %) in HBSS (1x), with phenol red Cat. No.: TRED1-100ML

#### General Information

Trypsin-EDTA solutions are used to detach adherent cells from culture surfaces. They are composed of natural porcine pancreas-derived trypsin and EDTA. The addition of phenol red serves as pH indicator to adjust to the perfect cell culture conditions.

The concentration of trypsin necessary to dislodge cells from their substrate is dependent primarily on the cell type and the age of the culture. Various formulations should be tested to determine the best product for a specific application.

Appearance	Clear frozen liquid
Storage and shelf life	Store at $\leq -15^{\circ}\text{C}$ . Avoid repeated freeze-thaw cycles. Preparation of aliquots recommended. Once opened, store at $4^{\circ}\text{C}$ and use within 2-4 weeks.
Shipping conditions	Frozen (Dry ice)
Thawing	$+37^{\circ}\text{C}$ water bath or overnight at $+2^{\circ}\text{C}$ to $+8^{\circ}\text{C}$ . Swirl gently to homogenize.

#### Formulation

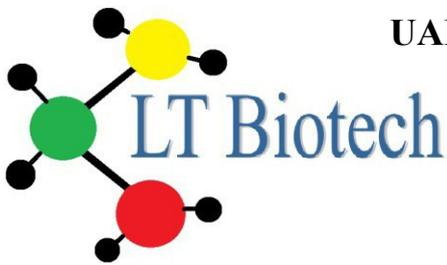
Components	Concentration
	mg/l
EDTA 2Na 2H <sub>2</sub> O	340.00
D-Glucose	1000.00
KCl	400.00
KH <sub>2</sub> PO <sub>4</sub>	60.00
NaCl	8000.00
NaHCO <sub>3</sub>	350.00
Na <sub>2</sub> HPO <sub>4</sub>	48.00
Phenol red	10.00
Trypsin	2500.00

#### Instructions for Use

##### *Detachment of adherent cells using Trypsin-EDTA*

Trypsin-EDTA (0.25 %) in HBSS (1x) with phenol red solution is supplied as a sterile, ready-to-use, frozen liquid. This entire procedure should be done in a laminar flow hood using proper aseptic technique.

1. The product can either be thawed in a  $+37^{\circ}\text{C}$  water bath or overnight at  $+2^{\circ}\text{C}$  to  $+8^{\circ}\text{C}$ .



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2. Carefully aspirate all of the media from the cell culture flask.
3. Rinse cells with  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$ -free salt solution (see related products), aspirate, and discard.
4. Prewarm the trypsin solution in a  $+37^{\circ}\text{C}$  water bath. Add enough trypsin solution to completely cover the cells.
5. Incubate the flask at  $+37^{\circ}\text{C}$ , or for more sensitive cultures, at room temperature or  $+2^{\circ}\text{C}$  to  $+8^{\circ}\text{C}$ .
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. Trypsin can cause cellular damage and time of exposure should be kept to a minimum.  
  
The time required to detach cells from the culture surface is dependent on the cell type, the age of the culture, population density, serum concentration in the growth medium and time since last subculture.
7. Neutralize trypsin 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.

### Precautions and Disclaimer

This product is for research use only.