



### Product Profile

Product Name:	Trypsin EDTA Solution C(0.05%) EDTA(0.02%)with Phenol Red
Catalog Number	03-053-1
Unit Size Availability:	(A) 500ml;(B) 100ml
Formulation:	Frozen Solution
Defined Storage Conditions:	-20°C
Stability: (Under Defined Handling & Storage Conditions)	Please Refer To Product Label

**Important Note!** Please read the MSDS and Product Profile carefully in their entirety before using this material for possible safety precautions and potential hazards.

#### Product Description:

Trypsin, an animal-derived product, is the most commonly used enzyme for harvesting cells in culture. Trypsin is a pancreatic serine protease (proteolytic) enzyme with specificity for peptide bonds involving the carboxyl group of the basic amino acids, Arginine and Lysine. Trypsin often contains a crude mixture of lipases, nucleases, polysaccharides and proteases extracted from Porcine pancreas.

Most cell cultures grow as a single thickness cell layer or sheet attached to a substrate known as a monolayer. When subculturing adherent cells, these intercellular and cell-to-substrate links or connections must be gently dissociated. Proteolytic enzymes such as Trypsin (i.e. a serine peptidase), breaks or gently separates these bonds by creating a single-cell suspension from which new subcultures are realized. Trypsin solutions are widely utilized as cell dissociation reagents for continuous cell culture of adherent growing cells. Trypsin proteolysis or trypsinization is a process in which proteins have been digested or treated with Trypsin and are thus said to be trypsinized. *Biological Industries'* Trypsin is designed not only to gently dissociate cells from almost any support substrates but also as well as from each other in order to actualize cell manipulation techniques in addition for other studies that require intact cell-surface proteins. Trypsin, as a solution, is available in a varied array of formulations with or without EDTA. EDTA is a chelator that binds calcium and magnesium ions that may otherwise inhibit the Trypsin activity which then hydrolyzes and gains access to the intercellular bonds being cell-cell and/or cell-substrate bonds. Crude Trypsin is often the subculturing agent of choice for cell dissociation/dissaggregation of adherent cells although the treatment may be cytotoxic if prolonged. Over-trypsinization is a common cause of subculture problems.

The use of Crude Trypsin often involves multiple changes and the variability among lots can dramatically influence the effectiveness of the dissociation process. Regarding the use of Crude Trypsin, some important facts must be noted:

- ✓ Cells must **NEVER** remain in the Crude Trypsin for longer than 3-5 minutes as they may be seriously damaged in the process (i.e. damage to the intracellular proteins).
- ✓ Cells should **NEVER** be left without a fluid layer
- ✓ Do not permit prolonged growth on culture ware as the cells will be very difficult to remove (i.e. after 5-7 days)

In a serum-free culture environment, the cells must be separated by rapid centrifugation or by utilizing Trypsin inhibitors such as Soybean Trypsin Inhibitor (SBTI). SBTI is a single polypeptide that forms a stable, stoichiometric, enzymically inactive complex with Trypsin thereby reducing the availability of Trypsin by, more or less, binding chymotrypsin. However, with *Biological Industries'* Soybean Trypsin Inhibitor, any excess Crystalline Trypsin may be completely neutralized, thereby avoiding the use of serum for this purpose. The cells may then be resuspended successfully in a suitable growth medium.

#### Predominant Characteristics of Trypsin EDTA Solution C (0.05%), EDTA (0.02), with Phenol Red include:

- § Animal-Derived Source
- § Trypsin(0.05%);EDTA(0.02%)
- § Meets USP and EP Testing Specifications
- § Cell-Culture-Tested
- § Suitable for Cell-Culture Applications
- § Long-Storage When Handled Properly Under Defined Conditions

#### Instructions /Procedures

- 1) Take a bottle from specified storage conditions of -20°C and read the Label. The Trypsin should be warmed to Room Temperature(15-30°) prior to use
- 2) Ensure that the cap of the bottle is tight.
- 3) Gently swirl the bottle to ensure homogeneity
- 4) Wipe the outside of the bottle with a disinfectant solution such as 70% Ethanol
- 5) Use Sterile/Aseptic Techniques in a laminar flow culture hood

**Storage & Stability:**

This product should be stored under specified conditions of -20°C and used within 18 Months. Do **not** use after the expiration date as specified on the label. Deterioration of liquid media may be recognized by any of the following characteristics, among others including: (a). color change, (b). granulation/ clumping, (c). insolubility,(d). And/or decrease in expected performance parameters. Any material described above should not be used and therefore discarded. Do not expose to light for prolonged periods as it is light-sensitive. For prolonged storage, store in the dark. **Quality Control\***(Please Note That Each Batch/Lot Will Differ as to the Final Specifications)

Test	Specifications*
Appearance:	Clear,Colorless
Cell Culture Test:	Test & Record
Cell Line:	Vero
Osmolality:	272-292 mOsm/kg
pH:	7.35-7.50
Sterility:	Sterile
Mycoplasma(According to USP)	Negative
Porcine Parvovirus:	Negative

*Conformity According to Guidance on Minimizing the Risk of Transmitting Animal Spongiform Encephalopathy Agents via Human and Veterinary Medical Products (EMA/410/01-Rev.1)*

**Production:**

*According To The Current European Pharmacopoeia: The Animals From Which Trypsin/Chymotrysin Is Derived Fulfill The Requirements For The Health Of Animals Suitable For Human Consumption To The Satisfaction Of The Suitable Authority; Furthermore The Tissues Used Do Not Include Any Specific Risk Materials Defined By Relevant International Legislation. The Method Of Production Allows Inactivation And Removal Of Any Contamination By Viruses And/Or Any Other Infectious Agents. The Method Of Manufacturing Is Validated.*

*We Hereby Confirm That The Trypsin/Chymotrysin Supplied By Biological Industries Is Not Manufactured From Raw Materials Whose Origins Present TSE/BSE Risk And Furthermore Does Not Contain Compounds Manufactured From Raw Materials With Origins Presenting Any TSE/BSE Risk.*

**Auxiliary Products**

Product Name	Product Concentration	Catalog Number	Unit Size	Storage Temperature
Trypsin Solution A With Calcium And Magnesium Without Phenol Red	0.25%	03-045-1B	100ml	2-8°C
Trypsin Solution B Without Calcium And Magnesium Without Phenol Red 10x Concentrate	2.50%	03-046-5A	500ml	-20°C
		03-046-5B	100ml	-20°C
Crystalline Trypsin Solution Without Phenol Red	0.02%	03-047-1A	500ml	-20°C
		03-047-1B	100ml	-20°C
Soybean Trypsin Inhibitor 50x Conc., 5mg/MI		03-048-1C	20ml	-20°C
Trypsin EDTA SolutionA EDTA(0.02%)	0.25%	03-050-1A/B	500ml/100ml	-20°C
Trypsin EDTA, EDTA 0.2% , 10X Conc.	0.50%	03-051-5B	100ml	-20°C
		03-051-5C	20ml	-20°C
Trypsin EDTA Solution B EDTA (0.05%) With Phenol Red	0.25%	03-052-1A	500ml	-20°C
		03-052-1B	100ml	-20°C
<b>Note:</b> For a list of Antibiotics, Serum, other Reagents and Supplements, please refer to our Product Catalog, Product Profiles, Product Guides and Internet Site.				

**References:**

- 1) Current Edition Merck Index
- 2) Biological Industries (BI) Specifications
- 3) Current Edition USP/E Ph
- 4) Martindale The Extra Pharmacopoeia, 28<sup>th</sup> Edition, Royal Pharmaceutical Society: London, England
- 5) Darling, D. C. and Morgan, S. J. Animal Cells: Culture and Media, John Wiley & Sons: New York, 1994