Product Name

Name: Trypsin Solution B (0.25%), without Calcium and Magnesium, without Phenol Red

Cat. No.: C3538-0100, C3538-0500

Size: 100 mL, 500 mL

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 two 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-cell thick layer or a 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), break or gently separate 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. VivaCell's 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 to 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. Trypsin is often the subculturing agent of choice for cell dissociation/disaggregation of adherent cells although the treatment may be cytotoxic if prolonged. Over-trypsinization is a common cause of subculture problems.

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

- Cells must NEVER remain in the trypsin solution for longer than 3-5 min as they may be seriously damaged in the process (i.e., damage to the extracellular proteins).
- Cells should NEVER be left without a fluid layer as cells will dry up very quickly.
- Do not permit prolonged growth (i.e., after 5-7 days) on culture-ware as the cells will be very difficult to dissociate from each other.

For serum-free culture experiments, the cells must be separated from the solution by immediate centrifugation or by utilizing trypsin inhibitors such as Soybean Trypsin Inhibitor (SBTI). SBTI is a single polypeptide that forms a stable, stoichiometric, and enzymatically inactive complex with trypsin thereby reducing the availability of trypsin by, more or less, binding chymotrypsin. However, with VivaCell's Soybean Trypsin Inhibitor, any excess crystalline trypsin may be completely neutralized, thereby without leaving any active trypsin behind. The cells may then be resuspended successfully in a suitable growth medium.

Predominant Characteristics

- Animal-derived source
- Meets USP and EP Testing Specifications
- Cell culture performance tested
- Suitable for cell culture applications
- Long-term storage when handled properly under defined conditions

Storage and Stability

The product should be kept at **2 - 8°C.**
The product is **light-sensitive** and therefore should not be left in the light.
Shelf life: 18 months from date of manufacture.

Procedure

- Take using a T25 culture flask as an example, during cell passage, aspirate the medium in the culture flask, add 3 mL PBS or DPBS (without calcium and magnesium) to rinse the cells, and then aspirate.
- Add 1 mL Trypsin Solution B to infiltrate the entire bottom surface, and put the flask into a 37°C incubator to digest for 3 - 5 min (adjust the specific digestion time according to the characteristics of the cell type).
- When most of the cells come off the bottom of the dish, mix with 3 mL complete medium, centrifuge at 200 - 250 x *g* for 3 min, and aspirate the supernatant.
- Resuspend the cells with a complete medium and passage as needed.

Quality control

Trypsin Solution B (0.25%), without Calcium and Magnesium, without Phenol Red is tested for sterility, pH, osmolality, mycoplasma. In addition, each batch is tested for cell growth performance.

Precaution and Disclaimer

For research use only, not for clinical diagnosis, and treatment.