Product Name

Name: Cell Dissociation Solution (non-enzymatic)

Cat. No.: C3505-0100

Size: 100 mL

Product Description

Most cell cultures grow as single-cell thickness or a cell sheet attached to a substrate known as a monolayer. When culturing adherent cells, these intercellular and cell-to-substrate links or connections must be gently dissociated. Normal attachment, growth and development of many cell types are dependent on attachment factors and extracellular matrix components. Attachment factors are structural proteins that have adherent capabilities and increase cell-substrate interactions in a culture-dependent attachment milieu. Well-known proteolytic enzymes, such as trypsin, gently break or separate these interactions by creating a single-cell suspension from which new subcultures are made. These proteolytic enzymes or serine peptidases are widely utilized as cell dissociation reagents for continuous cell culture or adherent growing cells. Although trypsin is an essential product for cell culture manipulation and is purified from animals, it is experiencing ever-increasing regulatory scrutiny especially in the biopharmaceutical manufacturing field. Therefore, there was a need to develop a protein-free, non-animal source trypsin, and alternative products for cell culture due to the potential of untoward contamination, such as viruses, other adventitious agents and/or unwanted enzymes.

Tissue dissociation, primary cell isolation and cell harvesting are some of the major applications for utilization of proteolytic enzymes (trypsin in particular) not only in cell culture in particular but also in cell biology research as a whole. In spite of their widespread use, their mechanisms of action (MOA's) are not well understood and therefore, the choice of one enzyme or technique over another is often empirical and arbitrary. Maximizing the yield of functionally viable, dissociated cells are often dependent upon, but not limited to the following parameters:

- Type of tissue
- Species
- Age
- Genetics
- Dissociation medium utilized
- Crude enzyme impurities
- Enzyme concentrations utilized
- Temperature
- Incubation times

Non-Enzymatic Cell Dissociation Solutions (CDS) are often employed when it is necessary to harvest cells by gently dislodging adherent cells from culture vessels especially when non-protein and animal-component free materials are the order of the day. CDS, a proprietary mixture of chelators, is a good alternative to animal-source proteins, proteolytic enzymes, and proteases such as collagenases, pronases and a wide array of trypsin formulations used for cell dissociation/disaggregation. The presence of certain proteases may interfere and/or modify cell membranes and cellular proteins causing untoward

manifestations in physiological or immunological assays.

This non-enzymatic CDS helps not only maximize the yield of functionally viable cells from culture vessels without the often untoward and cytotoxic effects of enzymes, but also with has the advantage that the cells may be exposed for longer periods of time without the negative ramifications of digestive enzymes and over-trypsinization which is a common cause of subculture problems. Cells can be dissociated easily from plastic or glass culture vessels and from each other with the CDS treatment. Another prominent advantage is that it is less labor-intensive, not subject to such lot/batch variability or the worry of the presence of other traces of interfering enzymes. However, this solution is not recommended for cell lines with very high adherent properties.

Predominant Characteristics

- Ready-to-use, chemically defined, non-enzymatic proprietary mixture of chelators
- Non-animal source alternative to trypsin without its untoward cytotoxicity
- Effective with serum-free or with serum-containing medium
- Meets USP and EP testing specifications
- Suitable for cell-culture & molecular biology applications
- Long-term storage when handled properly under defined conditions

Storage and Stability

The product should be kept at **2 - 8°C**.
Shelf life: 24 months from date of manufacture

Procedure

1. Pre-warm Non-Enzymatic Cell Dissociation Solution to 37°C.
2. Drain and discard the medium from the culture vessel without drying out the monolayer.
3. Rinse the monolayer with approximately 2.0 mL of Dulbecco's Phosphate Buffered Saline (DPBS) without Calcium and Magnesium or Non-Enzymatic Cell Dissociation Solution.
4. Add approximately 1.5 mL Non-Enzymatic Cell Dissociation Solution to the 25 mL culture vessel and gently swirl the vessel to completely bathe the monolayer.
5. Incubate cells at 37°C by periodically observing cells under a microscope until they begin to round up. Tapping the side of the vessel will help dislodge more adherent cells from the surface.
6. After detachment, disperse cells into suspension by repeated pipetting.
7. Centrifuge the cells at 200 x *g* for 2 - 5 minutes. Remove as much Non-Enzymatic Cell Dissociation Solution as possible and re-suspend the pellet with an appropriate medium.

Quality Control

Cell Dissociation Solution (non-enzymatic) is tested for sterility, pH, osmolality. In addition, each batch is tested for cell growth performance.

Precaution and Disclaimer

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