Non-Enzymatic Cell Dissociation Solution

30-2103™

Description

Trypsin is the most commonly used enzyme for harvesting cells in culture. A non-enzymatic approach is needed when non-protein and animal-component free materials are required. ATCC Non-Enzymatic Cell Dissociation Solution is a sterile, phenol-red free solution composed of a proprietary mixture of chelators representing an optimized alternative to protein-digesting enzymes. This product is totally free of animal-derived components. This product has applications for cell culture, cell growth, and viability.

Volume: 100 mL

Storage Conditions

Storage conditions: 2°C to 8°C

Intended Use

This product is intended for laboratory research use only. It is not intended for any animal or human therapeutic use, any human or animal consumption, or any diagnostic use.

BSL 1

ATCC determines the biosafety level of a material based on our risk assessment as guided by the current edition of *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, U.S. Department of Health and Human Services. It is your responsibility to understand the hazards associated with the material per your organization's policies

and procedures as well as any other applicable regulations as enforced by your local or national agencies.

Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at www.atcc.org.

Handling Procedures

Each type of cell or cell line responds to Non-Enzymatic Cell Dissociation Solution in a unique manner. For optimum results, frequently observe the cells during the dissociation process to prevent damage. For cell-specific information, please refer to the product sheet supplied with the cells or cell line.

Note: The use of Non-Enzymatic Cell Dissociation Solution is not recommended for highly adherent cell types, such as keratinocytes.

1. Bring D-PBS to room temperature before use. Warm the Non-Enzymatic Cell Dissociation Solution and complete growth medium to 37°C prior to use with the cells.
2. For each vessel, carefully aspirate the spent media without disturbing the monolayer. The type of rinsing agent to use is dependent on the composition of the complete growth medium; proceed with one of the following options.
    - Option 1: If the cell culture medium contains serum, each flask should be rinsed with D-PBS twice prior to adding the Non-Enzymatic Cell Dissociation Solution.
    - Option 2: If working with serum-free medium, each flask should be rinsed twice with 1 mM EDTA in D-PBS prior to adding the Non-Enzymatic Cell Dissociation Solution.
3. Using 1.5 mL for every 25 cm^2, add the appropriate volume of Non-Enzymatic

Cell Dissociation Solution to each vessel (e.g., each T-25 vessel would be dissociated with 1.5 mL Non-Enzymatic Cell Dissociation Solution).

4. Gently rock each flask to ensure complete coverage of the Non-Enzymatic Cell Dissociation Solution over the cells.
5. Place the flask(s) in a 37°C, 5% CO_2, incubator.
6. Observe the cells every 5 to 10 minutes under the microscope. When the cells pull away from each other and round up, remove the flask from the microscope and gently tap the culture vessel from several sides to promote detachment of the cells from the flask. (Some cell types may require more vigorous tapping.)
7. When the majority of cells appear to have detached, disperse the cells into suspension by repeated pipetting.
8. Transfer the dissociated cells to a sterile centrifuge tube and set aside while processing any remaining cells in the culture vessel.
9. Add 3 to 5 mL D-PBS to the tissue culture vessel to collect any additional cells that might have been left behind.
10. Transfer the cells and D-PBS to the centrifuge tube containing the Non-Enzymatic Cell Dissociation Solution-dissociated cells.
11. Repeat steps 8 and 9 as needed until all cells have been collected from all vessels.
12. Centrifuge the cells at 125 x g for 5 to 10 minutes.
    a. Do not over-centrifuge cells as this may cause cell damage.
    b. After centrifugation, the cells should form a clean loose pellet.
13. Aspirate neutralized dissociation solution and resuspend the cell pellet in 2 to 8 mL fresh, pre-warmed, complete growth medium.
14. Count the cells and seed new culture vessels at the recommended density.
15. Place newly seeded vessels in a 37°C, 5% CO_2, incubator, and incubate for at least 24 to 48 hours before processing the cells further.

Quality Control Specifications

Mycoplasma contamination: Not detected

Osmolality: 280 to 300 mOsm/kg

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: Non-Enzymatic Cell Dissociation Solution (ATCC 30-2103)

References

References and other information relating to this material are available at www.atcc.org.

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