



EnVzyme[™] Easy

Animal Component-Free Cell Dissociation Reagent For easy-to-dissociate cells Vegetable origin Sterile filtered

Product Code: TCL137

Product Description:

EnVzymeTM cell dissociation reagents are ready to use animal component-free reagents used as an alternative to trypsin for dissociation of adherent cells. These reagents are formulated to contain a proteolytic enzyme isolated from vegetable origin. Depending on their ability to dissociate different types of cells, they are available in two forms –

- EnVzymeTM Easy: For easy to dissociate cells
- EnVzymeTM Super: For hard to dissociate cells

Features

- Animal component-free: Derived from vegetable source. Eliminates the risk of viruses, BSE or other potential adventitious agents
- Gentle on cells: Gentle dissociation of cells without affecting the viability
- No inhibition required: EnVzymeTM reagents do not require addition of trypsin inhibitor for inactivation, hence they can be used conveniently with serum-free cultures.
- Retention of marker expression: Being gentle on the cells, EnVzymeTM reagents keep the cell surface epitopes unaltered. This feature makes them useful for surface marker expression analysis methodologies such as flow cytometry and immunocytochemistry^{*}.

TCL137 is EnVzymeTM Easy suitable for dissociation of easy-to-dissociate cells.

Directions:

- 1. Remove the spent medium from the culture vessel by aspiration.
- 2. Wash the monolayer by adding balanced salt solution without calcium and magnesium to the side of the flask opposite the cells.
- 3. Rinse the cell sheet by rocking the flask for 1 to 2 minutes and discard the wash solution.

4. Add EnVzymeTM Easy solution to the side of the flask opposite the cells. The volume should be sufficient enough to completely cover the monolayer of the cells. *Note: Refer Table 1 for quantity of EnVzymeTM to be used for different culture vessels.*

| Table 1: | EnVzymeTM | quantity | for | different | culture |
|----------|-----------|----------|-----|-----------|---------|
| vessels | | | | | |

| Culture vessel | Quantity of EnVzyme TM (µl) | | |
|----------------|--|--|--|
| 96-well plate | 50 | | |
| 48-well plate | 100 | | |
| 24-well plate | 100 - 200 | | |
| 12-well plate | 200 - 300 | | |
| 6-well plate | 400 | | |
| T12.5 flask | 300 | | |
| T25 flask | 500 | | |
| T75 flask | 1000 | | |
| T175 flask | 2000 | | |

5. Rock the flask to ensure that the dissociation solution covers the cell sheet.

- 6. Incubate the flask at 37°C and monitor the dissociation periodically by observing the flask under the microscope.
- 7. When dissociation is complete, the cells will be in suspension and appear rounded.

Note: In addition to rocking gently, the flasks may be tapped to expedite removal.

The exact time needed to dissociate cells will vary according to the cell line, cell density and time since last subculture. The dissociation process should be monitored closely to avoid cell damage.

8. Once the cell dissociation is complete add required quantity of medium.

- 9. Disperse the cells into a single cell suspension by pipetting repeatedly.
- 10. Count and seed the cells into fresh medium.

Quality Control:

Appearance

Colorless, clear solution.

pН

7.20 - 7.80

Cell lines tested:

Osmolality in mOsm/Kg H₂O

260.00 - 300.00

Sterility

No bacterial or fungal growth is observed after 14 days of incubation, as per USP specification.

Cell Dissociation Test

Maximum time required for complete dissociation of cells grown in T12.5 culture flask at 70 – 80% confluence: HUVEC: 7 min CHO: 5 min

| | | EnVzyme TM Easy | | |
|-----------|----------------------|---|--|--|
| Cell line | Culture Medium | Approx. time for complete dissociation | | |
| BHK-21 | MEM + 10% FBS | (min.)* 3 – 4 | | |
| CHO K1 | MEM + 10% FBS | 2-3 | | |
| L929 | DMEM + 10% FBS | 1 – 2 | | |
| HEK293 | DMEM + 10% FBS | 1 – 2 | | |
| WJ-MSC | MSC expansion medium | 1 – 2 | | |

* These are the approximate values of time required for complete dissociation of cells grown in T12.5 culture flask at 70 - 80% confluence. The duration may slightly vary depending on the culture vessel used, cell density (or the confluence) and time since last subculture.

Comparison of cell dissociation reagents:

| Specifications | EnVzyme TM Easy / EnVzyme TM Super | Trypsin / Trypsin- EDTA | Accutase® | ZymeFree TM |
|---|---|----------------------------|---------------|---------------------------------|
| Origin | Vegetable origin (Non-animal) | Animal origin | Animal origin | Chemically defined (Non-animal) |
| Storage | -20°C | -20°C | -20°C | 2 – 8°C |
| Inhibition required | Not required | Trypsin inhibitor | Not required | Not required |
| Activity at temperature | 37°C | 37°C | 37°C | 37°C |
| Compatibility with serum-free cultures | Compatible | Compatible | Compatible | Compatible |
| Compatibility with stem cells and primary cells | Compatible | Compatible | Compatible | - |

Storage and Shelf Life:

Shipping temperature: -20°C

Upon receipt store the product at -20°C in a freezer that is not self-defrosting.

Once thawed, the product is stable for about 2 weeks at 2-8°C.

Repeated freezing and thawing reduces enzymatic activity and should be avoided.

Once thawed, the solution can be aliquoted in smaller volumes and frozen for future use.

Use before expiry date given on the product label.

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Disclaimer:

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