Typical Cell Passaging Using Accutase®

Protocol





Accutase® is an effective solution for routinely detaching cells from standard tissue culture-treated vessels, as well as advanced surface treatments or coatings. Accutase is useful for cell detachment and for preparing single-cell suspensions from clumped cell populations for sub-culturing cells, analytical studies, and for accurate cell counting. Accutase is free of mammalian or bacterial-derived products, which reduces the risk of contamination.

Accutase is formulated at a concentration that is ready to use, once thawed. (Note: Never thaw Accutase at 37°C.) A thawed bottle of Accutase can be removed from the refrigerator and immediately applied to cells. It should not be pre-warmed to 37°C. Accutase contains proteolytic activity that gently breaks down cellular adhesion molecules and enables cell detachment from the bottom of the flask.

This following procedure should be performed in a laminar flow hood using proper aseptic technique.

- 1. Carefully aspirate all of the media from the cell culture flask. A subsequent rinse is not required.
- 2. Immediately add a volume of Accutase that is sufficient to cover the cells. This is typically 2.5 to 5 mL for a T-25 flask depending upon confluency and density of the culture.
- 3. Incubate the flask at room temperature for 5 to 10 minutes or up to a maximum of 1 hour. Check the flask frequently to confirm that the cells have rounded.
- 4. Once the cells have rounded, hit the flask against the palm of your hand to dislodge cells.
- 5. Gently disperse the cells by pipetting up and down, and then analyze the cell suspension to determine the viable cell density.
- 6. Add an aliquot of the cell suspension to fresh media in the desired number of new flasks, and incubate at 37°C. No neutralization steps are required. Cell attachment will occur within a few minutes, depending on cell type.

Warranty/Disclaimer: Unless otherwise specified, all products are for research use only. Not intended for use in diagnostic or therapeutic procedures. Not for use in humans. Corning Life Sciences makes no claims regarding the performance of these products for clinical or diagnostic applications.





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At Corning, cells are in our culture. In our continuous efforts to improve efficiencies and develop new tools and technologies for life science researchers, we have scientists working in Corning R&D labs across the globe, doing what you do every day. From seeding starter cultures to expanding cells for assays, our technical experts understand your challenges and your increased need for more reliable cells and cellular material.

It is this expertise, plus a 160-year history of Corning innovation and manufacturing excellence, that puts us in a unique position to offer a beginning-to-end portfolio of high-quality, reliable cell culture consumables.

For additional product or technical information, please visit www.corning.com/lifesciences/media or call 1.800.235.5476.

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