

OptiFRZ

Guidelines for Use

OptiFRZ®

DMSO-Free Cryopreservation Media
Animal-Origin-Free (AOF) and Chemically Defined
Made in USA

Introduction

Cryopreservation is a crucial tool that is utilized in many different applications across the life sciences field to preserve important cells and tissues. The need for a stable final cell product to be delivered to the patient is essential to ensure product efficacy and safety with cellular therapies. Traditionally, high inclusion levels of DMSO, a common cryoprotectant, are used to protect and maintain the integrity of the cells, however significant side effects from DMSO toxicity are often observed.

OptiFRZ has been engineered without the inclusion of DMSO while still providing the necessary nutrients required for cryopreservation. It is chemically defined and animal-origin-free, meaning there are no human or animal-derived components included. Further, all reagents used have previously been shown safe for injection.

Storage

Recommended storage at 2-8°C, tightly sealed, and protected from light.

Instructions for Use

Freezing:

1. Collect desired cell suspension for cryopreservation and obtain a cell pellet by centrifugation.
2. Carefully remove the supernatant and resuspend in cold OptiFRZ Media to obtain a cell suspension of $\sim 1 \times 10^6$ cells/mL.
3. Transfer 1 mL of the cell suspension directly into each cryovial.
4. Freeze cells using a controlled cooling rate protocol to achieve approximately $-1^\circ\text{C}/\text{minute}$.
5. After 24 hours, transfer cells to liquid nitrogen (-135°C) for long-term storage.

Recovery, Wash, and Counting:

1. To thaw, place samples in a 37°C water bath. Remove immediately when there appears to be no frozen cell pellet.
2. **Wash to remove OptiFRZ media** by transferring thawed cell suspension to a tube and diluting 10x with pre-warmed culture media or DPBS + 1% recombinant albumin ([Exbumin](#)® is recommended).
3. Centrifuge to obtain a cell pellet.
4. Remove supernatant and rewash cells 1-2 more times using the same pre-warmed culture media or DPBS + 1% recombinant albumin.
 - a. **Note: Cells must be washed at least 2 times to remove all traces of OptiFRZ media and ensure accurate viability assessment results**
5. Remove supernatant and resuspend with a minimal volume of pre-warmed culture media.
6. Pull a sample for viability assessment.
7. Plate in appropriate culture conditions evaluate post-recovery functionality.