

6042 Cornerstone Court W, Ste B San Diego, CA 92121

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# Cell Freezing Medium Catalog #: 79796-2 100 ml

### **Description**

Cryopreservation is a cornerstone of cell biology, and is essential to maintaining healthy and reliable cell lines. BPS Bioscience Cell Freezing Medium is optimized for maximum viability and rapid recovery of all BPS Bioscience Cell Lines. This product contains DMSO. It is provided sterile-filtered and mycoplasma-free for the protection of the samples.

# **Application**

Cryopreservation of mammalian cell lines

#### Quality

Sterile filtered (0.2 µm) and Mycoplasma-free. All components are low endotoxin. Serum is ISIA-certified and sourced from the United States of America.

### **Storage and Stability**

Store in aliquots at -20°C for up to 1 year. Avoid freeze/thaw cycles. Thaw overnight at 4°C before use. Hygroscopic; avoid moisture.

## **Cell Freezing Protocol**

- 1. If using adherent cells, rinse with phosphate buffered saline (PBS), and detach cells from culture vessel with 0.25% Trypsin/EDTA. Add growth medium to neutralize trypsin, and transfer to a conical tube.
- 2. Mix cells well by pipetting. Take a sample of the cells in a microcentrifuge tube, and count the number of viable cells by trypan blue staining. Calculate the number of cells available for freezing.



Note: For best results, cells should be at least 80% viable and preferably >90% viable before freezing, and in healthy log-phase growth, not overgrown.

- 3. Transfer cells from growth flask to a conical tube, and spin down cells for 5 minutes. Resuspend in 4°C BPS Bioscience Freezing Medium at ~2 x 10<sup>6</sup> cells/ml.
- 4. Dispense 1 ml of cell aliquots into cryogenic vials. Place vials in an insulated container for slow cooling (-0.5°C to -1°C per minute) and freeze at -80°C overnight.
- 5. Transfer the cells to liquid nitrogen the next day for long term storage.



Note: It is recommended to expand cells and freeze down more than 10 vials of cells for future use at an early passage.



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# **Cell Thawing Protocol**

- 1. Remove cells from liquid nitrogen storage and immediately put the vials on dry ice before thawing.
- 2. Warm Thaw Medium in a 37°C water-bath until temperature equalizes. (See cell line datasheet for medium recipes\*)
- 3. Thaw the frozen cells from dry ice in a 37°C water-bath for 40 seconds, until freezing medium is partially thawed but with some ice remaining.
- 4. Transfer the entire contents of the cell vial to a conical tube containing 10 ml of prewarmed Thaw Medium.
- 5. Spin down the cells for 5 minutes, remove the supernatant, and resuspend the cells in 5 ml of pre-warmed Thaw Medium.
- 6. Using a pipette, gently transfer the resuspended cells to a T25 flask and incubate at 37°C in a 5% CO<sub>2</sub> incubator.
- 7. After 24 hours of culture, change to fresh Thaw Medium for adherent cells, or add an additional 3-4 ml of Thaw Medium to suspension cells.
- 8. After cells reach relatively appropriate density, passage the cells using the protocol in the datasheet, and begin using selective antibiotics or Growth Medium.



\*Note: Thaw Medium does not contain selective antibiotics (G418, Hygromycin, Puromycin, etc.) because these can be too harsh on the recently thawed cells. We recommend that the selection be added back to cells after cells have recovered fully.

#### **Related Products**

| Product name                                   | Catalog# | Size   |
|--|----------|--------|
| Cell Staining Buffer                           | 79795    | 500 ml |
| ONE-Step™ Luciferase Assay System              | 60690-1  | 10 ml  |
| ONE-Step™ Luciferase Assay System              | 60690-2  | 100 ml |
| Dual Luciferase (Firefly-Renilla) Assay System | 60683-1  | 10 ml  |
| Dual Luciferase (Firefly-Renilla) Assay System | 60683-2  | 100 ml |
| Thaw Medium 1                                  | 60187-1  | 100 ml |
| Thaw Medium 2                                  | 60184-1  | 100 ml |
| Thaw Medium 3                                  | 60186-1  | 100 ml |
| Growth Medium 1A                               | 79528    | 500 ml |
| Growth Medium 2A                               | 60190    | 500 ml |
| Growth Medium 3A                               | 60188    | 500 ml |