

## **Product Information**

Cell Freezing Medium Serum Free

Catalog no: GBCFMSF01/01F

#### **General Information**

A serum-free cryoprotective media called Cell Freezing is based on an organic preservation medium that contains 10% DMSO. Its unique formulation assures good cell viability, making it particularly ideal for applications in stem cell research, including mesenchymal and hematopoietic stem cells, adult and embryonic stem cells, and induced pluripotent stem cells (IPS).

Applications include the high viability cryopreservation of a variety of cell types, ready-to-use solutions, cell banking, and stem cell storage.

### **Specification**

Appearance	Clear liquid
Storage and shelf life	Store at +2°C to +8°C. CFM is a light sensitive solution.
	It should be protected from light during shipping and storage.
Shipping conditions	Ambient

### **Freezing Procedure:**

Cells need to be examined for contamination before cryopreservation. Any conventional freezing procedure can be utilized using cell freezing media.

# For Suspension Culture.

- 1. Determine the number of cryopreserved viable cells. The middle of the log phase of cell development is ideal. To pellet the cells, centrifuge the cells for 5 minutes (200 to 400 g). As little supernatant as possible should be removed without disrupting the cells.
- 2. Re-suspend cells at a concentration of 5x106 to 1x107 cells/ml in pre-cooled (+4°C to +8°C) CFM.
- 3. Aliquot into vials for cryogenic storage. Vials should be placed at +4°C and the freezing process should begin immediately. By using programmed coolers or by putting vials in an insulated box in a freezer that is between -70°C and -90°C, cells are gently frozen at a rate of +1°C/min.
- 4. After that, move the storage vials to the storage of liquid nitrogen.

### **For Adherent Culture:**

- 1. Use a moderate dissociating agent to separate the cells from the substrate. Use Accutase\* (Cat. No. ACC-1B), especially with sensitive cells, to prevent cell damage. If required, deactivate the dissociating agent.
- 2. Establish the viable cell count after resuspending the detached cells in full growth media.



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- 3. To pellet cells, centrifuge for five minutes (200 to 400 g). As little supernatant as possible should be removed without disrupting the cells.
- 4. Re-suspend cells at a concentration of 5x106 to 107 cells/ml in pre-cooled (+4°C to +8°C) CFM.
- 5. Aliquot into vials for cryogenic storage. Vials should be placed at  $+4^{\circ}$ C and the freezing process should begin immediately. By using programmed coolers or by putting vials in an insulated box in a freezer that is between  $-70^{\circ}$ C and  $-90^{\circ}$ C, cells are gently frozen at a rate of  $+1^{\circ}$ C/min.
- 6. After that, move the storage vials to the storage of liquid nitrogen.

# The methods listed below can be used to defrost cryopreserved cells:

## Centrifugation:

- 1. Take the cells out of storage and promptly defrost them in a water bath at +37 °C. Genexis biotech advises utilising authorised safety eyewear for eye protection. We also advise wearing protective gloves to cover any exposed flesh.
- 2. Add 1 to 2 ml of defrosted cells to 25 ml of full growth media. Gently stir the cell suspension.
- 3. Centrifuge the cells for 2 to 3 minutes at 80 g.
- 4. Verify the supernatant's clarity and the pellet of a consolidated cell's visibility. Without disturbing the cells, discard the supernatant.
- 5. Completely resuspend the cells in growth media, then count the viable cells.

Plate the cells, step 6. At least 3x105 live cells should be used as the inoculum.

# **Plating Directly:**

- 1. Take the cells out of storage and promptly defrost them in a water bath at +37 °C. Genexis biotech advises utilising authorised safety eyewear for eye protection. We also advise wearing protective gloves to cover any exposed flesh.
- 2. Use full growth media to plate cells directly. For every 1 ml of frozen cells, use 10 to 20 ml of full media. At least 3x105 cells should be used as the inoculum.
- 3. Grow cells for 12 to 24 hours. To get rid of cryopreservative remnants, replace the medium with new complete growth media.

Particularly when treating delicate cells, we advise using step 1 of the defrosting process.

### Need Help?

If you have any further queries, please feel free to email our cell culture specialists at info@genexisbiotech.com



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#### Disclaimer:

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