

# PRODUCT DATA SHEET

## Cell Freezing Media

### Description

Atlanta Biologicals' Cell Freezing Media are a cost-effective alternative to in-house freezing preparations, and are designed for optimum protection and preservation of cells during frozen storage. These ready-to-use freezing media are suitable for the consistent cryopreservation of a broad spectrum of mammalian cells. Each lot of Cell Freezing Medium is quality tested to assure maximum performance.

Cell Freezing Medium–I contains DMEM 4.5 gm/l D-glucose, 20% Fetal Bovine Serum and 10% dimethyl sulfoxide. This cryopreservation medium is commonly used for hardy cell lines that are less susceptible to freezing damage.

Cell Freezing Medium–II contains 90% Fetal Bovine Serum and 10% dimethyl sulfoxide. This cryopreservation medium is typically used for sensitive cell lines since it offers more cryoprotection than Cell Freezing Medium-I.

### Product Use

**Cell Preparation:** Actively growing, healthy cell cultures should be used in the freezing procedure. This is best accomplished by maintaining cells in their log-phase of growth. For B-cell hybridomas, the preferred cell density is  $4\text{-}5 \times 10^5$  cells/ml with a viability exceeding 70%. Centrifuge the appropriate volume of cells (200xg 5-10 minutes), discard the culture supernatant and resuspend the cell pellet in cold (4°C-8°C) Cell Freezing Medium. A final cell density in the freezing medium of  $1\text{-}10 \times 10^6$  cells/ml is recommended. Transfer 1-2ml of the cell suspension into labeled cryovials. Allow the cells to equilibrate in the freezing medium at 4°C-8°C for 5-10 minutes - including your pipetting time.

**Cell Freezing:** Optimum recovery of viable cells following freezing is best accomplished by freezing the cells at an appropriate cooling rate. This may be accomplished using a low temperature freezer (-70°C) by placing the vials in a styrofoam box overnight and transferring the vials into liquid nitrogen storage in the morning. Alternatively, a programmable freezing unit may be used to freeze the cells at 1°C per minute until the cell suspension reaches a temperature of -30°C and then at 5-20°C per minute until the temperature reaches -100°C. The vials are then transferred to a liquid nitrogen freezer for long-term storage.

**Cell Thawing:** Rapidly thaw each vial of cells in a 37°C water bath. Transfer the contents to a 10X volume of growth medium and centrifuge the cell suspension at 200xg for 5-10 minutes. After discarding the supernatant, resuspend the cells in fresh growth medium at a final cell density of  $2\text{-}4 \times 10^6$  viable cells/ml and transfer the suspension into a cell culture flask.

*NOTE: Cells that are sensitive to osmotic shock should be diluted slowly (3-5 minutes) with fresh medium immediately after thawing and before centrifugation.*

*This product is manufactured for research and development purposes only. It is not intended for any human or animal diagnostic, therapeutic or other clinical uses. It is also not for agricultural, food, drug, cosmetic or household use. The use of these products must be supervised by a person technically qualified to handle potentially hazardous material.*

## PRODUCT DATA SHEET - Cell Freezing Media

### Precautions

The Cell Freezing Media is hazardous. Review the Material Safety Data Sheet for additional information before handling this product.

### Storage and Handling

The Cell Freezing Media is supplied in gamma irradiated, sterile PETG or PETE bottles. We recommend that the Cell Freezing Media be stored frozen at a temperature of -5°C to -20°C. The medium may be frozen and thawed several times; however, we recommend that multiple freeze-thaw cycles be avoided. If multiple freeze-thaw cycles are anticipated, the product may be aliquoted into sterile bottles for single-use applications. Cell Freezing Media can be stored for short periods at 2°-8°C without affecting performance. We recommend that this product be used within 12 hours after thawing or else refrozen. Always use aseptic techniques when handling the Cell Freezing Media.

### Shipping

The Cell Freezing Media is shipped frozen by next day air in insulated containers packed with dry ice.

*This product is manufactured for research and development purposes only. It is not intended for any human or animal diagnostic, therapeutic or other clinical uses. It is also not for agricultural, food, drug, cosmetic or household use. The use of these products must be supervised by a person technically qualified to handle potentially hazardous material.*