CELLBIO™
in service of your CELL

Technical Datasheet

CryoXL™ Cell Freezing Medium Glycerol, 1X

With FBS and Glycerol
Without Antibiotics and Phenol red
Sterile filtered

Product Code: TCL078

Product Description:

HiMedia's Cell Freezing Media are complete, ready to use reagents designed to protect and preserve cells during frozen storage. These media are a convenient and cost effective alternative to in-house freezing media and can be used for a wide variety of mammalian cells. Cell Freezing Medium with Glycerol is fully supplemented formulation prepared in Dulbecco's Modified Eagle Medium. It contains 10% Glycerol and fetal bovine serum. Glycerol acts as a cryoprotectant as it prevents formation of ice crystals and prevents cell damage. It does not contain phenol red. It is ready to use and does not require further addition of any other reagents.

This cryopreservation medium can be used for hardy cell lines that are less susceptible to freezing damage. Users are advised to test the susceptibility of the medium for sensitive cell lines.

Composition:

Ingredients	%V/V
OTHERS	
Dulbecco's Modified Eagle Medium	Made to volume
Fetal bovine serum	Proprietary
Glycerol	10%

Directions:

Cell freezing medium can be used with standard freezing protocols. The following protocol may be used. Thaw cell culture freezing medium, mix well, and keep on wet ice during use.

Procedure for freezing:

1. For optimum results, cells should be in log phase of growth.
2. Gently detach adherent cells from the surface using Trypsin or other appropriate means.
3. Gently pellet the cell suspension by centrifugation (200 to 400 x g for 5 minutes for suspension cells and 200 x g for 5 minutes for adherent cells). Using a pipette, remove the medium above the pellet down to the smallest volume without disturbing the cells.
4. Resuspend the cells in Cell Freezing Medium at a recommended density for a specific cell type. Hybridoma cells may require higher cell density.
5. Aliquot cells in appropriate cryogenic storage vials. Freeze the cells in a controlled rate freezing apparatus, decreasing the temperature approximately 1ºC per minute. Alternatively, place the cryovials containing the cells in an isopropanol chamber and store them at -80ºC overnight. Alternatively, store them at -20ºC for 1 - 2 hours before shifting to -80ºC overnight.
6. Transfer cryovials to liquid nitrogen tank for long term storage.

Procedure for thawing of cryopreserved cells:

1. Remove cells from frozen storage and quickly thaw in a 37ºC water bath.
2. Dilute 1ml suspension with 10ml of complete growth medium.
3. Mix cells gently and pellet by gentle centrifugation.
4. Discard the supernatant and gently resuspend the cells in complete growth medium and seed in appropriate culture vessel.
5. It is recommended to do viability assessment 24hours post thawing. For accurate assessment, it is recommended to use fluorescent assay like EZBlue™ assay (CCK004 - EZBlue™ Cell Assay Kit) or metabolic assays like MTT assay (CCK003-EZcount™ MTT Cell Assay Kit).

Notes:

1. Cells harvested for cryopreservation should be at their optimum viability to ensure maximum survival during freezing and after thawing.
2. On removal from storage, extreme caution must be exercised to prevent explosion of the cryovial because of sudden expansion of the trapped nitrogen.
3. To retain maximum viability during cryopreservation, cells must be cooled at a constant slow rate, -1 to -5ºC/min. This can be achieved using programmable freezers or placing ampoules in a heavily insulated box at -80ºC for 24 hours before transferring them to their final storage location.

4. After thawing cells, it is necessary to slowly dilute the croprotectant to prevent osmotic shock. When it is necessary to centrifuge the cells, use the minimum g force to sediment them to prevent shearing damage, i.e. 70-100g.
5. To initiate rapid growth, it is advisable to inoculate new cultures at a higher density than for routine subculture, e.g., between 3 and 4 X 10^4 viable cells/cm^2 for adherent cells.
6. The minimum number of tests that should be carried out on master cell banks are, total and viable cell counts, growth potential, screening for bacteria, fungi and mycoplasma and cell line authenticity.

Quality Control:

Appearance
Pale yellow colored clear solution.

pH
7.60 - 8.20

Sterility
No bacterial or fungal growth is observed after 14 days of incubation, as per USP specification.

Post-thaw viability of BHK-21
85%

Post-thaw viability of CHO
85%

Performance Test
Performance test is done by freezing cells and doing a viability assessment after thawing and comparing it with a control medium.

Storage and Shelf Life:

Cell Freezing Media should be stored at -30°C to -10°C.
For frequent use, cell freezing medium once thawed can be stored at 2-8ºC up to 5 days.
The shelf life of product is 12 months.
Use before expiry date given on the product label.

Disclaimer :

Revision : 05/2025

User must ensure suitability of the product(s) in their application prior to use. Products conform solely to the information contained in this and other related HiMedia™ publications. The information contained in this publication is based on our research and development work and is to the best of our knowledge true and accurate. HiMedia™ Laboratories Pvt Ltd reserves the right to make changes to specifications and information related to the products at any time. Products are not intended for human or animal or therapeutic use but for laboratory, diagnostic , research or further manufacturing use only, unless otherwise specified. Statements contained herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.