

## Best-in-class optimized biopreservation media for cells and tissues

- Pre-Formulated
- Serum-Free
- Protein-Free
- USP/Highest Quality Components
- cGMP Manufactured
- FDA Master File
- Sterility, Endotoxin, and Cell-Based Release Testing



CryoStor®, a series of cell-specific, optimized freeze media, is designed to prepare and preserve cells in ultra low temperature environments (-70°C to -196°C). CryoStor®, pre-formulated with DMSO, provides a safe, protective environment for cells and tissues during the freezing, storage, and thawing process. Through modulating the molecular-biological response to the cryopreservation process, CryoStor® provides for enhanced cell viability and functionality while eliminating the need for serum, proteins, or high levels of cytotoxic agents.

#### Glossary of label symbols:

REF	LOT	STERILE	
Part Number	Lot Number	Aseptic Processing Technique	
EXP. Date	2°C 4°C Temperature	MFG. By	MFG. Date



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 BioLifeSolutions.com

### Ordering Information

Product Name	Size	Part #
CryoStor® CS2	100mL bottle	202102
CryoStor® CS5	100mL bottle	205102
CryoStor® CS5	100mL bag	205202
CryoStor® CS5	10mL vial	205373
CryoStor® CS10	100mL bottle	210102
CryoStor® CS10	100mL bottle	210502*
CryoStor® CS10	100mL bag	210202
CryoStor® CS10	10mL vial	210373
CryoStor® CS10	16mL vial	210374
CryoStor® CS10	1000mL bag	210210

\*New bottle type - contact BioLife Solutions for details

### To Order

**Call:** 1.866.424.6543 | **Fax:** 425.402.1433  
**Sales:** SalesOne@BioLifeSolutions.com  
**Web:** BioLifeSolutions.com  
**Technical Support:** info@BioLifeSolutions.com

# CryoStor® CS2, CS5 and CS10 Freeze Media

## Usage and Cryopreservation Protocol

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1. Place cells to be cryopreserved into suspension (mechanical or enzymatic dissociation).
2. Centrifuge cells to obtain cell pellet.
3. Remove supernatant - Note: Remove as much culture media as possible, to reduce dilution of CryoStor solution.
4. ISOLATION: Add cold (2°-8°C) CryoStor.
  - a. Cell concentrations: 0.5-10 × 10<sup>6</sup> cells/mL for routine cell culture protocols (higher [cell] possible).
  - b. DMSO is pre-mixed in CryoStor - no additives are necessary.
5. PRE-FREEZE: Incubate cell suspension at 2°-8°C for approximately 10 minutes.
6. NUCLEATION: Freeze samples at -80°C (many protocols utilize -70°C and -80°C interchangeably).
  - a. Use a controlled rate freeze (-1°C/min) or similar protocol for most mammalian cell systems.
  - b. The freezing device or isopropanol container should be pre-cooled to 2°-8°C.
  - c. Ice nucleation within the sample (seeding) should be initiated at approximately -5°C using either a liquid nitrogen burst program setting on a controlled rate freezer or mechanical agitation (flick or tap) of the cryovial/sample container after approximately 15-20 min. at -80°C.
  - d. Freeze time (-80°C) using isopropanol containers is recommended to be approximately 4 hours, or not more than overnight.
7. STORAGE: Place samples into storage.
  - a. Store samples at liquid nitrogen temperatures (below -130°C).
  - b. Sample storage at -80°C is only recommended for short-term storage (weeks to months).
8. THAWING: Thaw samples quickly in a 37°C water bath, or equivalent mechanical thawing device.
  - a. Sample thawing should be conducted with gentle swirling of sample until all visible ice has melted. Approximate thaw time for a 1 ml sample in a cryovial is approximately 2-3 minutes.
  - b. DO NOT allow sample to warm above chilled temperatures (0-10°C). Cryovials should be cool to the touch when removed from bath. Passive thaw is not recommended.
9. Dilute cell/CryoStor mixture immediately with culture media, or equivalent isotonic media.
  - a. Dilution procedure can be performed in a single step.
  - b. The dilution media should be between 20°C and 37°C.
  - c. A dilution ratio of 1:10 (sample to media) or greater is recommended.
10. Plate cells in appropriate configuration.
11. Place cells into culture conditions or utilize immediately.
12. Viability assessment 24 hours post-thaw\*.

Note: To obtain an accurate measure of cell viability following cryopreservation, assessment should be performed 24 hours post-thaw and compared to non-frozen controls.

*\*Sample assessment immediately post-thaw with membrane integrity indicators, such as Trypan Blue, for comparative analysis of sample cell yield and viability often results in inaccurate measurement of cell survival.*

Live/Dead fluorescent assays or metabolic assays (MTT or alamarBlue®) are recommended for more accurate assessment of viable recovery.

CryoStor products ship at ambient temperature. Upon receipt, store at 2°-8°C, protected from light, until ready to use.

Further protocol support is available at [info@BioLifeSolutions.com](mailto:info@BioLifeSolutions.com).

### Materials are manufactured under cGMP

Test methods and criteria are provided on all lot specific Certificates of Analysis and Release.