

CryoStor® CS2, CS5 and CS10

Freeze media

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



BEST-IN-CLASS OPTIMIZED BIOPRESERVATION MEDIA FOR CELLS AND TISSUES

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.

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 bag	210202
CryoStor® CS10	10mL vial	210373
CryoStor® CS10	16mL vial	210374
CryoStor® CS10	1L bag	210210
CryoStor® CS10	10mL syringe	210473

To Order

Call: 1.866.424.6543 | **Fax:** 425.402.1433

Sales: salesone@BioLifeSolutions.com

Web: BioLifeSolutions.com

Technical Support: info@BioLifeSolutions.com

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Usage and Cryopreservation Protocol

- 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: $0.5-10 \times 10^6$ 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.

Materials are manufactured under cGMP

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