

CAPTIVATE BIO

GUIDELINES FOR USE

CRYOVATE™ SF Freezing Medium

Product Description

Captivate Bio's CRYOVATE™ SF Freezing Medium is a 2X, serum-free and animal component-free cryopreservation media containing dimethyl sulfoxide (DMSO).

- Animal-free, protein-free, and chemically defined solution
- Serum and phenol red free
- Preserves viability, attachment, and cell growth
- Prepared with ACS grade (Research Grade) components
- Manufactured in the US in ISO-certified facilities

CRYOVATE™ SF Freezing Medium | Cat. No. CRY014 | 50 mL
Store at -20°C for up to 24 months from date of manufacture

Refer to the Safety Data Sheet (SDS) for hazard information.

Materials and Reagents

- CRYOVATE™ SF Freezing Medium (2X)
- Conditioned or serum-free medium
- Clean bench or biosafety cabinet
- Sterile pipettes and pipette tips
- Sterile tubes and cryogenic vials
- Freezing container or device
- Liquid nitrogen tank or -80°C freezer for storage
- Additional: water bath at 37°C, fridge, and centrifuge

Directions for use

1. Wipe the outside of the CRYOVATE SF bottle with 70% ethanol or isopropanol.
2. Dilute the 2X formulation and create a 1:1 solution with either a conditioned medium or a fresh serum-free medium before use.
3. Harvest cells according to standard protocols ensuring high viability (>90%).
4. Resuspend cells in diluted 1:1 solution at the recommended cell density (typically 1-10 x 10⁶ cells/mL).

Cryopreserving Cells

1. Label each of your cryogenic vials with cell line, passage number, date, and researcher initials.
2. In a biosafety cabinet, gently mix the cell suspension, avoiding bubbles, and ensure homogeneous distribution.
3. Gently pipette and dispense 1 mL of cell suspension into each cryovial.
4. Incubate cells at 2-8°C for 10 minutes.
5. Transfer and cryopreserve cells using slow controlled-rate freezing protocol (approximately -1°C minute) in an isopropanol or alcohol-free freezing container.
6. Freeze at -80°C overnight, then transfer vials to a liquid nitrogen tank.

NOTE: Long-term storage at -80°C is not recommended.

Thawing Cells

1. Warm cell culture medium of choice in a 37°C water bath.
2. Wipe the outside of your cryovials with 70% ethanol or isopropanol.
3. Quickly thaw cells in a 37°C water bath by gently swirling the vial. Do not submerge the vial. Remove the vial when only a small frozen cell pellet remains. Do not vortex cells.
4. Transfer the cell suspension to a sterile tube containing pre-warmed serum-free medium.
5. Centrifuge the cell suspension at 200-300 x g for 5 minutes to remove the cryo medium.
6. Repeat steps as needed.
7. Resuspend the pellet in fresh medium and plate. Cells are now ready for use in downstream applications.

For more information, visit [captivatebio.com](https://www.captivatebio.com), email orders@captivatebio.com, or contact us at (617) 607-4017.