# MesenCult™-ACF Freezing Medium



### Animal component-free MSC cryopreservation medium

Catalog # 05490 50 mL

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## **Product Description**

MesenCult<sup>™</sup>-ACF Freezing Medium is a defined, serum-free, and animal component-free medium for the cryopreservation of mesenchymal stem and progenitor cells (MSCs). This complete and ready-to-use medium is recommended for human MSCs previously cultured in MesenCult<sup>™</sup>-ACF Plus Medium (Catalog #05445) or MesenCult<sup>™</sup> Medium (MesenCult<sup>™</sup> Proliferation Kit; Catalog #05411). Frozen human MSCs should be stored at -135°C (liquid nitrogen) or colder.

- Defined, serum-free, and animal component-free
- Reproducibly high recovery rates
- Optimized for MSCs previously cultured in MesenCult™-ACF Plus Medium or MesenCult™ Medium
- Preserves human MSC multipotency and expansion capacities
- Convenient, ready-to-use format

## Properties

Storage: Store at 2 - 8°C.

Shelf Life: Stable until expiry date (EXP) on label.

Contains: Dimethyl sulfoxide (DMSO)

Please refer to the Safety Data Sheet (SDS) for hazard information. This product contains components dissolved in dimethyl sulfoxide (DMSO). DMSO is a strong solvent and skin penetrant, and can transport many substances through the skin. DMSO can also penetrate some protective glove materials including latex and silicone. Extra caution should be utilized when handling this product.

# Handling / Directions For Use

#### FRFF7ING

- 1. Wipe the outside of the MesenCult™-ACF Freezing Medium container with 70% ethanol or isopropanol before opening.
- 2. Prepare a single-cell suspension of human MSCs using the desired dissociation medium (e.g. Animal Component-Free Cell Dissociation Kit; Catalog #05426) and centrifuge cells at 300 x g for 5 minutes to obtain a cell pellet.
- 3. Carefully remove the supernatant with a pipette, leaving a small amount of medium to ensure the cell pellet is not disturbed. Resuspend the cell pellet by gently flicking the tube.
- 4. Add cold (2 8°C) MesenCult™-ACF Freezing Medium to obtain a cell suspension of 1 x 10^6 cells/mL and mix thoroughly.
- 5. Transfer 1 mL of the cell suspension into each cryovial.
- 6. Freeze cells using a standard slow rate-controlled cooling protocol (approximately -1°C/minute) or an isopropanol freezing container and store at -135°C (liquid nitrogen).
  - NOTE: Long-term storage at -80°C is not recommended.

#### **THAWING**

For complete instructions on culturing MSCs using a specific culture medium, please refer to the Product Information Sheet for the medium (e.g. MesenCult™-ACF Plus Medium; Document #DX22329).

NOTE: When serum-free medium is used, pre-coating of the cultureware is generally required. Cultureware should be prepared in advance according to the supplier's instructions and brought to room temperature (15 - 25°C) for at least 30 minutes prior to use.

- 1. Warm DMEM/F-12 with 15 mM HEPES (Catalog #36254) and desired culture medium (e.g. MesenCult™-ACF Plus Medium) in a 37°C water bath.
- 2. Wipe the outside of the vial of cells with 70% ethanol or isopropanol.
- 3. In a biosafety cabinet, twist the cap a quarter-turn to relieve internal pressure and then retighten.
- 4. Quickly thaw cells in a 37°C water bath by gently shaking the vial. Remove the vial when a small frozen cell pellet remains. Do not vortex cells.
- 5. Wipe the outside of the vial with 70% ethanol or isopropanol.

### MesenCult™-ACF Freezing Medium



- 6. Transfer cells to a 15 mL polypropylene tube (e.g. Catalog #38009) containing 10 mL of warm DMEM/F-12 with 15 mM HEPES.
- 7. Centrifuge cells at 300 x g for 5 minutes at room temperature (15 25°C).
- 8. Carefully remove the supernatant with a pipette, leaving a small amount of medium to ensure the cell pellet is not disturbed. Resuspend the cell pellet by gently flicking the tube.
- 9. Add 2 mL of culture medium to the tube. Mix gently.
- 10. Plate cells into pre-coated cultureware (e.g. T-75 cm² flask or 6-well plate).

  NOTE: Add 1.2 x 10^5 to 2.5 x 10^5 cells per T-75 cm² flask, or 15,000 to 40,000 cells/well of a 6-well plate.
- 11. Move the cultureware in several quick, short, back-and-forth and side-to-side motions to evenly distribute the MSCs across the surface. Place the cultureware in a 37°C incubator.

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