

# MesenCult™-SF Attachment Substrate



**Serum-free attachment substrate for human mesenchymal stem cells**

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Catalog # 05424

5 mg

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## Product Description

MesenCult™-SF Attachment Substrate is a serum-free substrate for the in vitro culture of human mesenchymal stem and progenitor cells (MSCs). The substrate supports both expansion of human MSCs in vitro as well as their enumeration using the colony-forming unit-fibroblast (CFU-F) assay.

## Properties

**Storage:** Store at 2 - 8°C.

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

This product contains material derived from human plasma. Donors have been tested and found negative for HIV-1 and -2, hepatitis B, and hepatitis C prior to donation. However, this product should be considered potentially infectious and treated in accordance with universal handling precautions.

## Handling / Directions For Use

1. Dissolve the lyophilizate with 5 mL sterile tissue grade water to obtain a final concentration of 1 mg/mL. Do not agitate or pipette vigorously.
2. Incubate at 37°C for 30 - 60 minutes to ensure the lyophilizate is fully in solution. Swirl gently.  
NOTE: If not used immediately, store solution at -20°C. It is recommended to aliquot the reconstituted solution into multiple vials (e.g. 500 µL). Avoid repeated freeze-thaw cycles.
3. Coat plates one day prior to use (i.e. coat overnight at 2 - 8°C), or, if time is limited, coat at room temperature (15 - 25°C) for 2 hours. It is recommended to coat plates overnight.
4. Plates must be coated with different dilutions of MesenCult™-SF Attachment Substrate when performing CFU-F assays and when expanding cells in culture, as follows:

### COATING PLATES FOR CFU-F ASSAY

Dilute MesenCult™-SF Attachment Substrate 1 in 30 in D-PBS (Without Ca<sup>++</sup> and Mg<sup>++</sup>; Catalog #37350). Gently mix. Prepare an amount slightly more than required to account for pipetting variability. For example, to coat one 6-well plate, dilute 160 µL of MesenCult™-SF Attachment Substrate in 4.8 mL of D-PBS.

### COATING PLATES FOR EXPANSION

#### When culturing cells obtained from primary tissue:

Dilute MesenCult™-SF Attachment Substrate 1 in 20 in D-PBS (Without Ca<sup>++</sup> and Mg<sup>++</sup>; Catalog #37350). Gently mix by inverting the tube twice. Prepare an amount slightly more than required to account for pipetting variability. For example, to coat one T-75 cm<sup>2</sup> flask, dilute 250 µL MesenCult™-SF Attachment Substrate in 5 mL D-PBS.

#### When culturing previously cultured cells:

Dilute MesenCult™-SF Attachment Substrate 1 in 28 in D-PBS (Without Ca<sup>++</sup> and Mg<sup>++</sup>; Catalog #37350). Gently mix by inverting the tube twice. Prepare an amount slightly more than required, to account for pipetting variability. For example, to coat one T-75 cm<sup>2</sup> flask, dilute 185 µL MesenCult™-SF Attachment Substrate in 5 mL D-PBS.

5. Add diluted MesenCult™-SF Attachment Substrate (step 4) as follows:
  - 6-well plate: 800 µL per well
  - T-25 cm<sup>2</sup> flask: 2 mL per flask
  - T-75 cm<sup>2</sup> flask: 5 mL per flask
6. Wrap plates with Parafilm®, sealing the junction between the base and lid and incubate at 2 - 8°C overnight, or at room temperature for 2 hours. For flasks, seal the vent on the cap with Parafilm® and incubate as described.

7. If plates/flasks were incubated overnight at 2 - 8°C, bring to room temperature (approximately 20 minutes) prior to washing. Gently pipette off remaining MesenCult™-SF Attachment Substrate without touching the newly coated surface.
8. Wash plates/flasks once with sterile distilled water by slowly pipetting water down the side of the well/flask, being careful not to scrape the coated surface. Swirl gently to rinse the entire surface, then carefully aspirate the water.
9. Allow to dry at room temperature for at least 15 minutes prior to use.

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