Product Name

Name: SpeeDiff MSC Adipogenic Differentiation Medium

Cat. No.: C8210-0100, C8210A-0001, C8210B-0001

Size: 100 mL

Product Description

SpeeDiff MSC Adipogenic Differentiation Medium is specifically designed to differentiate Mesenchymal Stem Cells (MSCs) of various origins into adipose tissue.

SpeeDiff's formulation accommodates MSCs previously cultured in serum-containing medium (e.g., FBS), serum-free medium (SFM, e.g., human platelet lysate), and/or animal component-free (ACF) mediums. MSCs from different origins—such as adipose tissue (AD), bone marrow (BM), and umbilical cord (UC)—can efficiently differentiate and form lipid droplets using this medium.

Special Features

- **High Induction / Differentiation Rate:**
  Depending on cell origin, passage number, and culture conditions, AD/BM-MSCs can be induced within 3–7 days, and UC-MSCs can be induced within 7–10 days.
- **Clear Morphology:**
  MSCs exhibit typical large, round lipid droplets post-induction. Oil Red O staining yields a high differentiation ratio with a clear background.
- **High Stability:**
  The fully formulated complete medium can be stored at 2–8°C, protected from light, for up to 2 months with minimal decay in efficacy.
- **Streamlined Protocol:**
  Unlike traditional adipogenic differentiation media, SpeeDiff MSC Adipogenic Medium is less toxic to MSCs. There is no need to alternate between differentiation and expansion media to rescue or cycle the cells during the induction phase. This single-medium protocol significantly shortens operation time and reduces costs.

Applications

This medium features an optimized composition to enhance lipid droplet formation and accumulation. It is thoroughly validated and manufactured in an ISO 13485 certified, GMP-compliant facility. Each lot is rigorously tested to guarantee performance and consistency, making it ideal for studies on adipose generation mechanisms, drug screening, regenerative medicine and MSC quality control (QC).

Components

Name (Cat. No.)	Volume (mL)	Appearance	Storage
SpeeDiff Basal Medium for Adipogenic Differentiation (C8210-0100)	100	Red transparent Liquid	-20℃
SpeeDiff Supplement I for Bone Marrow and Adipose-Derived MSCs (C8210A-0001)	0.2	Colorless transparent Liquid	-20℃
SpeeDiff Supplement II for Umbilical Cord Derived MSC (C8210B-0001)	0.2	Light Yellow transparent liquid	-20℃

Instructions for Use

1. Preparation of complete medium:

1.1 Place the basal medium and Supplement I or II at 2–8°C until completely thawed. Invert and tap the supplement tube, then briefly spin it down to collect all volume.

Note: Thawed basal medium may show precipitation due to its FBS content; this does not adversely affect performance. Precipitation can be removed by centrifuging at 400 × g for 3–5 minutes and transferring the supernatant to a new bottle. Removing flocculants via filtration is not recommended, as it may result in a loss of volume and active ingredients.

1.2 Add the entire volume of supplement (0.2 mL) to the basal medium (100 mL), mix thoroughly and allow the mixture to equilibrate to room temperature.

1.3 Tightly close the complete medium bottle, label

it with the name and date, and store it at 2–8°C away from light. Use within 2 months.

1.4 Aliquot the complete medium to avoid repetitive freeze-thaw cycles or temperature fluctuations.

2. MSC induction:

2.1 Reagent preparation:

Reagent	Function
MSC expansion medium	Normal MSC expansion
1xDPBS (VivaCell C3591)	Wash cell
0.25% Trypsin (VivaCell C3530)	Cell dissociation
SpeeDiff MSC Adipogenic Differentiation Complete Medium	To induce MSC adipose differentiation

2.2 MSC Induction Protocol (Example using a 12-well plate)

Note: We recommend to precoat culture plasticware by gelatin (VivaCell, C3260) to minimize cell edge retraction, detachment and floating during differentiation culture.

1) Add 0.3-0.5 mL, 0.1% gelatin in each well, shaking evenly to cover entire bottom surface.
2) Place the 12-well plate in a biosafety cabinet or laminar hood for at least 15 min.
3) Aspirate the gelatin solution; the plate is now ready for cell seeding.
4) Seed MSCs at a density of 2-3×10^4 cells/cm^2 per well and add 1 mL of MSC expansion medium.
5) Incubate the plate in a 37℃, 5% CO_2, 90% RH incubator.
6) When cell confluency reaches 80% - 90%, remove the expansion medium and add 1 mL of SpeeDiff MSC Adipogenic Differentiation Complete Medium.
7) Change the complete differentiation medium every 2–3 days. Increase the frequency of medium changes if the color turns yellow prematurely.
8) Repeat step (7) until sufficient adipose droplets are observed in the cells, indicating the cells are ready for staining.

3. Oil Red – O Staining:

Note: To prevent cell detachment, handle the plates gently, and perform all pipetting steps smoothly.

3.1 Remove the differentiation medium and gently wash each well 1–2 times with 1× DPBS.

3.2 Use the VivaCell Adipo-dye Staining Kit (VivaCell C37A0-0150). Prepare the Oil Red O working solution and centrifuge it at 400 × g for 5 minutes prior to use to lower the background staining.

3.3 Observe the stained lipid droplets under a microscope.

Note: Stained plates can be wrapped in Parafilm and stored at 4°C for up to 1 week. Prolonged storage may cause the oil droplets to merge or diffuse.

For Semi-Quantitative Analysis:

3.4 Aspirate the staining solution, add 1 mL of DPBS into each well, incubate at room temperature for 1 minute, and discard the supernatant.

3.5 Add a sufficient volume of 100% isopropanol (0.5–0.7 mL) to cover the entire surface of each well. Incubate at room temperature for 20 minutes. If intact oil droplets remain visible, prolong the incubation time until they are completely dissolved.

3.6 Measure the absorbance using a plate reader set to a wavelength of 510 nm.

Note: It is highly recommended to include a cell-free negative control well.

FAQ

- Why do the edges of the cell colonies curl or retract after induction?

Answers:

(1) Gelatin Coating: The culture surface was not precoated with gelatin.

(2) Cell Confluency: Confluency was too high at the start of induction. It is recommended to begin induction strictly at 80%–90% confluency.

(3) Temperature Shock: Thermal shock occurred between the medium and the cells. Ensure all reagents are equilibrated to room temperature before adding them to the cells.

- Why is the positive differentiation rate low, or taking longer than expected?

Answers:

(1) Cell Condition: The cell passage number may be

too high, culture conditions might be suboptimal, or the culture experienced pH fluctuations.

(2) Improper Reagent Mixing: Incomplete addition or inadequate mixing of the basal medium and supplement components.

- Why is the background filled with dark red debris after staining with Oil Red O?

Answers:

Centrifugation Step: The Oil Red O working solution was not centrifuged. Always centrifuge at 400 x g for 5 minutes after preparation and use only the supernatant.

Storage and Stability

- Dry ice shipment
- Storage: -10 ~ -20℃, protect from light
- Shelf Life: 12 months

Precautions

- Do not use it if the packaging is damaged, broken, or leaking.
- Maintain aseptic technique to avoid contamination during handling.
- Rinse immediately with tap water, if the solution contacts skin and mucous membrane.
- Always use the product before its expiration date.

Disclaimer

For research use only, not intended for clinical diagnosis, or human/animal treatment.