MesenCult™-ACF Chondrogenic Differentiation Medium

Animal component-free medium for the differentiation of MSCs into chondrocytes

Catalog #05455 100 mL

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Product Description

MesenCult™-ACF Chondrogenic Differentiation Medium is animal component-free and specifically formulated for the in vitro differentiation of human mesenchymal stem and progenitor cells (MSCs) into chondrogenic lineage cells, including chondrocytes. This medium is suitable for the differentiation of human bone marrow (BM)-, adipose tissue (AT)- and synovium (S)-derived MSCs previously culture-expanded in serum-containing medium (e.g. MesenCult™ Proliferation Kit [Human; Catalog #05411]) or serum- and animal component-free MesenCult™-ACF Plus Medium (Catalog #05445).

Product Information

The following components are sold as a complete medium (Catalog #05455) and are not available for individual sale.

COMPONENT NAME	COMPONENT #	SIZE	STORAGE	SHELF LIFE
MesenCult™-ACF Chondrogenic Differentiation Basal Medium*	05456	95 mL	Store at 2 - 8°C.	Stable for 12 months from date of manufacture (MFG) on label.
MesenCult™-ACF 20X Chondrogenic Differentiation Supplement	05457	5 mL	Store at -20°C.	Stable until expiry date (EXP) on label.

*Basal Medium stored for more than 2 months following the date of manufacture, as indicated on the label, should be supplemented with additional L-glutamine. For example, add 200 µL of 200 mM L-Glutamine (Catalog #07100) to 20 mL of Basal Medium to achieve a final concentration of 2 mM.

Preparation of Complete MesenCult™-ACF Chondrogenic Differentiation Medium

Use sterile techniques to prepare complete MesenCult™-ACF Chondrogenic Differentiation Medium (Basal medium + Supplement). The following example is for preparing 20 mL of complete medium. If preparing other volumes, adjust accordingly.

1. Thaw MesenCult™-ACF 20X Chondrogenic Differentiation Supplement at room temperature (15 - 25°C) or at 2 - 8°C overnight.

   NOTE: Once thawed, use immediately or prepare 1 mL aliquots and store at -20°C. Do not exceed the expiry date as indicated on the label. Once aliquots are thawed, use immediately. Do not re-freeze.
2. Add 1 mL of MesenCult™-ACF 20X Chondrogenic Differentiation Supplement to 19 mL of MesenCult™-ACF Chondrogenic Differentiation Basal Medium in a 50 mL polypropylene tube (e.g. Catalog #38010). Mix thoroughly.

   NOTE: Ensure the tube is polypropylene; do not use polystyrene or any other type of tube.

   NOTE: If not used immediately, store complete MesenCult™-ACF Chondrogenic Differentiation Medium at 2 - 8°C for up to 1 week. This medium does not contain antibiotics. If desired, add antibiotics and use medium within 1 week.

Directions for Use

3D PELLET CULTURE SYSTEM FOR CHONDROGENIC DIFFERENTIATION OF MSCs

The following example is for preparing 4 pellets. If preparing other amounts, adjust accordingly.

Day 0

1. Resuspend 2 x 10^6 MSCs in 2 mL of complete MesenCult™-ACF Chondrogenic Differentiation Medium at room temperature (15 - 25°C).
2. Add 0.5 mL of the cell suspension to each of 4 x 15 mL polypropylene tubes (e.g. Catalog #38009). Cap tightly and centrifuge at 300 x *g* for 5 - 10 minutes at room temperature (15 - 25°C).

   NOTE: Ensure the tubes are polypropylene; do not use polystyrene or any other type of tube.
3. Very gently loosen the cap of each tube (while still keeping the cap on with a half twist) and place in a rack.
4. Incubate at 37°C and 5% CO_2 for 3 days.

Day 3

5. Gently add 0.5 mL of complete MesenCult™-ACF Chondrogenic Differentiation Medium at room temperature (15 - 25°C) to each tube, for a final volume of 1 mL. Incubate tubes in the rack at 37°C and 5% CO_2 for 3 days.

Day 6 - 21

6. On Day 6 and every 3 days afterward, carefully aspirate the medium without disturbing the pellet and replace with 0.5 mL of complete MesenCult™-ACF Chondrogenic Differentiation Medium at room temperature (15 - 25°C). Incubate tubes in the rack at 37°C and 5% CO_2.

   NOTE: After each medium change, gently flick each tube to ensure the pellet is not completely attached to the tube.

   NOTE: The pellets may significantly increase in size throughout the incubation period.

Day 21

7. The chondrogenic pellets have reached full differentiation and can be used for downstream applications, or for quantitative and qualitative characterization analysis. Histological sections of the pellet can be generated by fixing the pellets in 10% formalin at room temperature (15 - 25°C) for 30 minutes, following subsequent standard paraffin embedding methods and staining 6 µm sections with Alcian Blue and Nuclear Fast Red.

Notes and Tips

- Do not use less than 0.5 mL of complete medium per pellet culture.
- It is recommended to change medium on a 3-day cycle. However, if the medium begins to turn yellow, switch to a 2-day cycle.