

MesenCult™ Osteogenic Differentiation Kit (Human)



Scientists Helping Scientists™ | [WWW.STEMCELL.COM](http://www.stemcell.com)

TOLL FREE PHONE 1 800 667 0322 • PHONE +1 604 877 0713

INFO@STEMCELL.COM • TECHSUPPORT@STEMCELL.COM

FOR GLOBAL CONTACT DETAILS VISIT OUR WEBSITE

Catalog #05465

1 Kit

Product Description

MesenCult™ Osteogenic Differentiation Kit (Human) is specifically formulated for the in vitro differentiation of human mesenchymal stem and progenitor cells (MSCs) into cells of the osteogenic lineage. This kit is suitable for the differentiation of human bone marrow (BM)-derived MSCs previously culture-expanded in serum-containing medium (e.g. MesenCult™ Proliferation Kit [Human; Catalog #05411] or MesenCult™-hPL [Human; Catalog #05439]) or serum-free medium (e.g. xeno-free MesenCult™-XF Medium [Catalog #05420] or animal component-free MesenCult™-ACF Medium [Catalog #05440]). MesenCult™ Osteogenic Differentiation Kit (Human) is also suitable for the differentiation of mesenchymal progenitor cells derived from embryonic stem (ES) cells or induced pluripotent stem (iPS) cells obtained by using STEMdiff™ Mesenchymal Progenitor Kit (Catalog #05240).

NOTE: MesenCult™ Osteogenic Differentiation Medium must be supplemented with L-Glutamine (Catalog #07100).

Product Information

The following components are sold as a kit (Catalog #05465) and are not available for individual sale.

| COMPONENT NAME | COMPONENT # | SIZE | STORAGE | SHELF LIFE |
|---|-------------|--------|-------------------|--|
| MesenCult™ Osteogenic Differentiation Basal Medium (Human) | 05466 | 200 mL | Store at 2 - 8°C. | Stable until expiry date (EXP) on label. |
| MesenCult™ Osteogenic Differentiation 5X Supplement (Human) | 05467 | 50 mL | Store at -20°C. | Stable until expiry date (EXP) on label. |

None of the above components contain antibiotics.

Preparation of Complete Osteogenic Differentiation Medium (Human)

Use sterile techniques to prepare complete Osteogenic Differentiation Medium (Basal Medium + 5X Supplement + L-Glutamine). The following example is for preparing 50 mL of complete medium. If preparing other volumes, adjust accordingly.

NOTE: For aliquoting and storing either the supplement or the complete medium, polypropylene tubes are strongly recommended (e.g. Falcon® Conical Tubes, 15 mL [Catalog #38009] or 50 mL [Catalog #38010]).

1. Thaw the 5X Supplement at room temperature (15 - 25°C) or at 2 - 8°C overnight. Mix thoroughly.

NOTE: Once thawed, use immediately or aliquot and store at -20°C. Do not exceed the shelf life of the supplement. After thawing the aliquoted supplement, use immediately. Do not re-freeze.

2. Add 10 mL of 5X Supplement to 40 mL of Basal Medium. Mix thoroughly.
3. Add 0.5 mL of L-Glutamine (200 mM; Catalog #07100) to reach a final concentration of 2 mM. Mix thoroughly.

NOTE: If not used immediately, store complete Osteogenic Differentiation Medium at 2 - 8°C for up to 1 week.

Directions for Use

Please read the entire protocol before proceeding.

For instructions on culturing human MSCs using the MesenCult™ media listed below, refer to the Product Information Sheets available at www.stemcell.com.

- MesenCult™ Medium (Human; Catalog #05411)
- MesenCult™-hPL Medium (Human; Catalog #05439)
- MesenCult™-XF Medium (Catalog #05420)
- MesenCult™-ACF Medium (Catalog #05440)

For differentiating to the osteogenic lineage, use culture-expanded human MSCs between passage 1 - 4.

The following protocol is for setting up differentiation assays using human BM-derived MSCs in a 6-well plate. If using other cultureware, adjust volumes accordingly.

NOTE: Only use tissue culture-treated cultureware.

1. Plate cells in 2 mL of growth medium per well. Recommended cell plating densities are as follows:
 - MesenCult™ Medium (Catalog #05411): $8 - 16 \times 10^3$ cells/cm²
 - MesenCult™-ACF Medium, MesenCult™-XF Medium, or MesenCult™-hPL Medium (Human): $4 - 8 \times 10^3$ cells/cm²NOTE: For MesenCult™-ACF and -XF, ensure to coat cultureware as described in the Product Information Sheets for those products.
2. Incubate at 37°C in 5% CO₂ with $\geq 95\%$ humidity until cells are approximately 90 - 98% confluent. This takes approximately 1 - 5 days depending on the expansion medium used.
3. Aspirate medium and replace with 2 mL of complete Osteogenic Differentiation Medium per well. Incubate at 37°C in 5% CO₂ with $\geq 95\%$ humidity.
4. Change medium every 3 - 4 days until bone matrix formation occurs (approximately 10 - 15 days).
5. Visualize osteogenic differentiation by staining with Alizarin Red S, alkaline phosphatase, or silver nitrate (von Kossa).

NOTE: The level of osteogenic differentiation for MSCs may vary depending on cell source, donor, and previous culture conditions.

STEMCELL TECHNOLOGIES INC.'S QUALITY MANAGEMENT SYSTEM IS CERTIFIED TO ISO 13485. PRODUCTS ARE FOR RESEARCH USE ONLY AND NOT INTENDED FOR HUMAN OR ANIMAL DIAGNOSTIC OR THERAPEUTIC USES UNLESS OTHERWISE STATED.

Copyright © 2017 by STEMCELL Technologies Inc. All rights reserved including graphics and images. STEMCELL Technologies & Design, STEMCELL Shield Design, Scientists Helping Scientists, and MesenCult are trademarks of STEMCELL Technologies Canada Inc. Falcon is a registered trademark of Corning Incorporated. All other trademarks are the property of their respective holders. While STEMCELL has made all reasonable efforts to ensure that the information provided by STEMCELL and its suppliers is correct, it makes no warranties or representations as to the accuracy or completeness of such information.