Mesenchymal Stem Cell Media

PromoCell

Instruction Manual

Mesenchymal Stem Cell Growth Medium

Product	Size	Catalog Number
Mesenchymal Stem Cell Growth Medium (Ready-to-use)	500 ml	C-28010

Mesenchymal Stem Cell Differentiation Medium

Product	Size	Catalog Number
Mesenchymal Stem Cell Adipogenic Differentiation Medium (Ready-to-use)	100 ml	C-28011
Mesenchymal Stem Cell Chondrogenic Differentiation Medium (Ready-to-use)	100 ml	C-28012
Mesenchymal Stem Cell Osteogenic Differentiation Medium (Ready-to-use)	100 ml	C-28013
Mesenchymal Stem Cell Chondrogenic Differentiation Medium w/o Inducers (Ready-to-use)	100 ml	C-28014
Mesenchymal Stem Cell Neurogenic Differentiation Medium (Ready-to-use)	100 ml	C-28015

Recommended for

- Human Mesenchymal Stem Cells from Bone Marrow (hMSC-BM)
- Human Mesenchymal Stem Cells from Umbilical Cord Matrix (hMSC-UC)
- Human Mesenchymal Stem Cells from Adipose Tissue (hMSC-AT)

Product Description

PromoCell Mesenchymal Stem Cell Media were developed for the *in vitro* expansion and directed differentiation of mesenchymal stem cells (MSC) from bone marrow, the umbilical cord matrix (Wharton´s Jelly) and adipose tissue.
PromoCell Mesenchymal Stem Cell Media are available as Medium (Ready-to-use) and consist of a bottle of Basal Medium and one vial of SupplementMix. Adding the SupplementMix to the Basal Medium results in the complete Medium.

Supplementation Details

PromoCell Mesenchymal Stem Cell Media contain all the growth factors and supplements necessary for the optimal expansion and directed differentiation of human mesenchymal stem cells.

Note: The PromoCell MSC Chondrogenic Differentiation Medium w/o Inducers (C-28014) must be supplemented with adequate chondrogenic inducers by the customer. The PromoCell MSC Chondrogenic Differentiation Medium (C-28012), the MSC Chondrogenic Differentiation Medium w/o Inducers (C-28014) and the MSC Neurogenic Differentiation Medium (C-28015) are serum-free and chemically defined.
PromoCell Mesenchymal Stem Cell Media do not contain antibiotics or antimycotics and are formulated for use in an incubator with an atmosphere of 5% CO_2.

Preparation of the supplemented Medium for Use

Thaw the SupplementMix in a 37°C water bath with occasional swirling. Do not incubate longer than necessary! In case of visible precipitates after complete thawing, mix gently until all precipitates have re-dissolved. Then, transfer the entire content of the SupplementMix to the Basal Medium. Close the bottle and swirl gently until a homogenous mixture is formed.

Instructions for the Use of PromoCell MSC Differentiation Media

Adipogenic Differentiation Protocol
1. Plate 6 x 10^4 MSC per well in a 24-well tissue culture plate (3.15 x 10^4 cells/cm^2) using MSC Growth Medium (C-28010). Work in duplicate.
2. Let the cells reach 80 - 90% confluency (24 - 48 hours).

3. Induce one of the duplicate samples with MSC Adipogenic Differentiation Medium (C-28011). Use MSC Growth Medium for the remaining well as a negative control.
4. Incubate for 14 days. Change Medium every third day. Be careful not to disturb the cell monolayer.
5. Optional: Fix and stain the lipid vesicles formed within the mature adipocytes.

Chondrogenic Differentiation Protocol
1. Plate 1 x 10^5 MSC per well in a 96-well U-bottom suspension culture plate using MSC Growth Medium (C-28010). Work in duplicate.
2. Spheroids will spontaneously form within 24 - 48 hours.
Note: The more cells are used, the bigger the spheroids (up to 3 x 10^5 cells per well).
3. Induce one of the duplicate samples with MSC Chondrogenic Differentiation Medium (C-28012). Use MSC Growth Medium for the remaining well as a negative control.
4. Incubate for 21 days. Change Medium every third day. Be careful not to aspirate the spheroids.
5. Fix and stain the cells for chondrogenic markers.

Osteogenic Differentiation Protocol
1. Plate 6 x 10^4 MSC per well in a 24-well tissue culture plate (3.15 x 10^4 cells/cm^2) using MSC Growth Medium (C-28010). Work in duplicate.
2. **Important:** Let the cells reach ≥100% confluency (24 - 72 hours).
3. Induce one of the duplicate samples with MSC Osteogenic Differentiation Medium (C-28013). Use MSC Growth Medium for the remaining well as a negative control.
4. Incubate for 21 days. Change Medium every third day. Be careful not to disturb the cell monolayer.
5. Fix and stain cells for osteogenic markers.

Neurogenic Differentiation Protocol
1. Coat an appropriate tissue culture vessel with fibronectin (C-43050) according to the instruction manual. Use a 10 µg/ml solution.
2. Plate 4,000 cells/cm^2 onto the fibronectin coated plate using MSC Growth Medium (C-28010). Work in duplicate.
3. Culture the cells to 80 - 90% confluency.
4. Induce one of the duplicate samples with MSC Neurogenic Differentiation Medium (C-28015). Use MSC Growth Medium for the remaining well as a negative control.
5. Incubate for at least 3 days. Change Medium every 48 hours.
6. Optionally, fix and stain for neuronal markers. Be careful not to disturb the cell monolayer.

For detailed information, please see www.promocell.com/application-notes.

Storage and Stability

Store the Basal Medium at 4 to 8°C in the dark and the SupplementMix at -20°C immediately after arrival. Do not freeze the Basal Medium. If stored properly, the products are stable until the expiry date stated on the label. After adding the supplements to the Basal Medium, the shelf life of the complete medium is 6 weeks at 4 to 8°C. Do not freeze the complete medium.
For use, pre-warm only an aliquot of the complete medium and keep the remaining medium refrigerated at 4 to 8°C.
Note: The SupplementMix is delivered thawed and can be frozen after arrival without losing any activity.

Quality Control

All lots of PromoCell Mesenchymal Stem Cell Media are subjected to comprehensive quality control tests using primary human mesenchymal stem cells. Each lot of MSC Growth Medium is checked for maintenance of multipotency, growth promoting activity, adherence rate, and typical morphology of the tested mesenchymal stem cells. Each lot of MSC Differentiation Media is tested for the capacity to induce directed differentiation into the respective lineages in MSC. Approved in-house lots of media are used as a reference.
In addition, all lots of media have been tested for the absence of microbial contaminants (fungi, bacteria, mycoplasma).

Intended Use

The products are for *in vitro* use only and not for diagnostic or therapeutic procedures. For safety precautions please see appropriate MSDS.

Note: PromoCell media are not suitable for trypsin neutralization (e.g. when splitting the cells). Instead we recommend using our DetachKit (C-41200, C-41210, C-41220), which contains HEPES BSS, Trypsin/EDTA and Trypsin Neutralizing Solution.

If you require special media modifications, we offer a Custom Media Service starting at 10 bottles per order. Please ask for details.

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04/2013