

Mesenchymal Stem Cell Medium Kit, xeno-free

Product Description

Mesenchymal Stem Cell Medium kit is a xeno-free, enhanced Mesenchymal Stem Cell medium. It consists of optimized Mesenchymal Stem Cell basal medium and a cell culture supplement which contains a human Platelet-derived growth factor mixture necessary for cell growth and proliferation of MSCs.

Intended use

For human *ex-vivo* tissue and cell culture processing applications, e.g. cell cultivation or cell isolation.

Protocols

Preparation of Complete Medium

1. Warm the basal medium, the supplement and antibiotics to 37°C.
2. Add supplements to the basal medium.
3. Keep growth medium at 37°C.
4. The supplemented medium can be stored at 4°C up to 6 weeks.

Cell Cultivation

1. Calculate seeding cell number for your cell culture flask (e.g.: T-25, seeding density: $5 \times 10^3/\text{cm}^2$; seed 125,000 per T-25).
2. Add 10 mL of xeno-free Mesenchymal Stem Cell medium to a T-25 flask.
3. Add appropriate number of cells to the cell culture flask. Cultivate in an incubator at 37°C and 5% CO₂.
4. Cells will attach within 2-4 hours after seeding. 70% confluence will be reached after 4-6 days.
5. Passage cells and sub-culture with a seeding density of $5 \times 10^3 - 1 \times 10^4$ cells/cm².
6. Change medium every two days.

Passaging Cells

1. Remove and discard xeno-free Mesenchymal Stem Cell medium from the T-25 flasks.
2. Wash cells twice with 10 mL HBSS.
3. Use warm (37°C) Trypsin-EDTA solution (1.5-2.0 mL) or our cell dissociation reagent CellSplit (1.0-1.5 mL) for a T-25 flask.
4. As soon as cells have detached (the flask may require a few firm gentle taps), add 10 mL of xeno-free Mesenchymal Stem Cell medium to the flask.
5. Transfer cells in a 15 mL tube and centrifuge at 200 xg for 5 minutes at room temperature.
6. Discard the supernatant.
7. Re-suspend cell pellet in a known volume (~ 1 mL) of xeno-free Mesenchymal Stem Cell medium.
8. Count the cells using Trypan Blue exclusion.

9. Seed cells with 5×10^3 - 1×10^4 cells/cm².
10. Add 10 mL of xeno-free Mesenchymal Stem Cell medium to a T-25 flask
11. Change medium every 2 days.

Cryopreservation Protocol

Material:

Phosphate Buffered Saline (PBS)
 Trypsin-EDTA solution or CellSplit
 Cell Culture Media
 Cryo-Media
 Labeled Cryovials (~ 3 per 100 mm plate)
 T-25 flask or T-75 flask confluent cells

Procedure:

1. Flush the adherent layer with a 5 mL sterile pipette 3-5 times to dislodge loosely attached cells.
2. Remove and discard the cell culture media from the flask.
3. Wash adherent cells 2-3 times with 10 mL of sterile PBS without calcium and magnesium to remove non adherent cells.
4. Remove and discard the wash solution from the flask.
5. Incubate cells with warm (37°C) Trypsin- EDTA solution or CellSplit for 1-3 minutes. Use 2.0-3.0 mL of Trypsin-EDTA solution or 3.0-4.5 mL of CellSplit when collecting cells from T-75 flasks and 1.5-2.0 mL of Trypsin-EDTA solution or 1.0-1.5 mL of CellSplit when using T-25 flasks. Prevent too long exposure to Trypsin-EDTA solution or CellSplit, as this may cause irreversible damage to the cells.
6. As soon as cells have detached (the flask may require a few firm gentle taps; please control detachment under the microscope), add 10 mL of xeno-free Mesenchymal Stem Cell medium to the flask.
7. Centrifuge the cell suspension at 200 x g for 5 minutes.
8. Remove supernatant with sterile Pasteur pipette.
9. Quickly re-suspend pellet by adding 1 mL freezing media per vial to be frozen.

Product Information

| Product Code | Description | Size | Shelf life |
|--------------|--|------|------------|
| MSC-001A | Mesenchymal Stem Cell Kit (500mL basal medium and 25mL MSC supplement) | Kit | 12 months |

Storage

This product is stable until the expiry date stated on the label. The Mesenchymal Stem Cell basal medium is most stable when stored frozen at +2 to +8°C. The recommended storage for the Mesenchymal Stem Cell Growth supplement is -20°C.

Once prepared, the fully supplemented (complete) medium may be stored for up to 6 weeks at +4°C. Warming of the media and repeated freeze/thaw cycles of the product should be avoided.

Recommendation

It is recommended that our product CellSplit, a xeno-free cell dissociation reagent be used. This is a product of vegetable origin and the active reagent is a substitute for Trypsin.

Safety Information

Follow the handling instructions outlined in the Safety Data Sheets (SDS). Wear appropriate protective eyewear, clothing and gloves. This is a Human origin material which has been tested as non-reactive at donor level for anti-HIV 1 & 2, anti-HCV and HBsAg. This product must be handled in accordance with established bio-safety practices.

Despite all testing, proper safety precautions for potentially infectious agents must be taken. All human blood products should be handled in accordance with currently acceptable bio-safety practices and guidelines for the prevention of blood borne viral infections.

Intended Use

This product is manufactured from outdated human platelet units concentrates and is intended for *in vitro* experimental and research use only. These products are sold for research and manufacturing purposes only. They are not intended for use in humans or animals.

Support

Life Science Production is a division of Life Science Group Ltd.
Life Science Production is [ISIA Traceability Certified](#)
Life Science Group Ltd is an ISO 9001:2015 Certified company

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