InnoCellular Tech Pte Ltd
Block 79 Ayer Rajah Crescent, #05-03, Singapore 139955
(E) enquiry@Innocellular.com | www.innocellular.com.sg

MesenPlify™ sXF

Product Sheet

Catalogue Number C0001

Product Description

MesenPlify™ sXF is a **Xeno-free, Coating-free, Serum-free** complete media formulation designed to support the *in-vitro* growth and proliferation of human mesenchymal stem cells (hMSCs). The use of MesenPlify™ sXF demonstrates exceptional capabilities in facilitating the long-term maintenance and proliferation of hMSCs, while preserving their multi-lineage differentiation potential (Osteogenic, Adipogenic, Chondrogenic) and essential cell surface expression markers (CD73, CD90, CD105) as defined by the ISCT minimal criteria.

MesenPlify™ sXF has been tested to be highly suitable for culturing a wide range of hMSCs, including Bone Marrow derived (BM-MSC), Adipose derived (AD-MSC) and Umbilical Cord derived (UC-MSCs).

Properties

MesenPlify™ sXF is supplemented with human platelet lysate (hPL) from human source, xeno-free alternative to Fetal Bovine Serum (FBS), which is frequently used for growth and expansion of cells. hPL has been shown to have better lot to lot consistency compared to FBS.

Components

The MesenPlify™ sXF cell culture kit consists of 2 components:

Product	Catalogue Number	Volume	Storage Condition	Shelf Life
MesenPlify™ sXF Basal Media	S0001	235mL	4°C	1 Year from date of manufacture
MesenPlify™ sXF Supplement	SU0001	15mL	-20°C	1 Year from date of manufacture

None of the above component contain antibiotics.

InnoCellular Tech Pte Ltd
Block 79 Ayer Rajah Crescent, #05-03, Singapore 139955
(E) enquiry@Innocellular.com | www.innocellular.com.sg

MesenPlify™ sXF

Product Sheet

Catalogue Number C0001

Materials Required but Not Supplied

Trypsin
D-PBS (without Ca++, without Mg++)
Cuture dish and flasks
Polypropylene conical tubes

Safety

Treat all human derived materials (e.g. hMSCs, hPL) as potentially infectious.

Read the product SDS, adhere to BSL-2 practises, wear appropriate personal protective equipment, handle the materials in a certified Biological Safety Cabinet.

Preparation of complete media

(Note: Perform all procedures using aseptic techniques.)

1. Thaw the MesenPlify™ sXF supplement (Catalogue No. SU0001) at room temperature or 2-8°C.
(Note: Use immediately after thaw, or aliquot in polypropylene tubes and free at -20°C. Do not use beyond the shelf life of individual components. Do not re-freeze aliquots.)

2. Add 15ml of MesenPlify™ sXF supplement to 235mL MesenPlify™ sXF Basal Media (Catalogue No. S0001). Mix thoroughly.

3. Store the complete MesenPlify™ sXF media at 2-8°C, away from light, for up to 2 weeks.
(Note: If desired, an additional filtration step using 0.2um filter units can be performed. If desired, antibiotics can be added at 1% of final complete media volume.)

Guideline for use

Recovery of Cryopreserved hMSCs

1. Pre-warm complete MesenPlify™ sXF media to 37°C.

2. Rapidly thaw a frozen vial of hMSC in a 37°C water bath until a small amount of ice remains.

3. Transfer the contents of the vial into a 50ml conical tube. Slowly pipet 10ml of complete MesenPlify™ sXF media dropwise into the conical tube.

MesenPlify™ sXF

Product Sheet

Catalogue Number C0001

4.Centrifuge at 300 x g for 5 minutes at room temperature.

5. Discard supernatant to remove DMSO cryoprotectant.

6. Resuspend the cell pellet in 1mL of complete MesenPlify™ sXF media

7. Perform cell count.

8. Top up cell suspension with complete MesenPlify™ sXF media to an appropriate volume. *(Recommendation: Use 30mL of MesenPlify™ sXF media in 150mm dish and 15mL for T75 flask.)*

9. Seed cells in tissue culture treated flasks at 3,000-5,000 cells per cm^2.
(Note: A higher seeding density is recommended when cells are first recovered from cryopreservation)

10. Maintain cells in a humidified CO_2 incubator at 37°C, 5% CO_2.

11. Perform complete media change every 3-4 days with complete MesenPlify™ sXF media pre-warmed to 37°C.

Sub-culture of hMSCs

1. Check that hMSCs are 70 - 80% confluent. Do not allow hMSCs to reach 100% confluency, which cells would exhibit signs of differentiation.

2. Pre-warm complete MesenPlify™ sXF media to 37°C

3. Gently wash cells once with 10mL of DPBS (without Ca++, without Mg++, not provided in the kit).

4. Add sufficient trypsin (not provided in the kit) to cover entire surface of tissue culture flask. *(Recommendation: Use 3ml of trypsin per T75 flask and 5ml of trypsin per 150mm dish).*

5. Swirl flask to spread trypsin across surface.

6. Incubate at 37°C for 2-10 mins.

7. Using a phase-contrast microscope, check for cell detachment. Gently tap the sides of the flask to aid in detachment.

8. Once cells have detached and are free-floating, add complete MesenPlify™ sXF media to 2x volume of trypsin.

MesenPlify™ sXF

Product Sheet

Catalogue Number C0001

(Recommendation: Use 6ml of complete MesenPlify™ sXF media *per T75 flask and 10ml of* complete MesenPlify™ sXF media *per 150mm dish).*

9. Collect cell suspension in a 50ml conical tube.

10. Centrifuge at 300 x g for 5 minutes at room temperature.

11. Discard supernatant to remove trypsin.

12. Resuspend the cell pellet in 1mL of complete MesenPlify™ sXF media.

13. Perform cell count.

(Note: Sample may need to be further diluted in media if cell density exceeds recommended range of cell counter.)

14. Calculate volume of cell suspension needed to seed at 3,000 - 5,000 cells per cm^2.

15. Top up cell suspension with complete MesenPlify™ sXF media to an appropriate volume. *(Recommendation: Use 30mL of MesenPlify™ sXF media in 150mm dish and 15mL for T75 flask.)*

16. Maintain cells in a humidified CO_2 incubator at 37°C, 5% CO_2.

17. Perform complete media change every 3-4 days with complete MesenPlify™ sXF media pre-warmed to 37°C.

Cryopreservation of hMSCs

1. Prepare 2X cryopreservation media by supplementing 20% of Dimethyl Sulfoxide (DMSO, not provided in the kit).

2. Resuspend cell pellet to twice of the desired cell concentration (e.g. 2 x 10^6 /mL) in pre-warmed complete MesenPlify™ sXF media.

3. Slowly add the 2X cryopreservation media (prepared in step 1) to the cell suspension, and gently mix by pipetting.

4. Transfer the cell suspension to pre-chilled (2°C - 8°C) cryovials.

5. Place the cryovials at -80°C in a cryogenic freezing container overnight.

6. Transfer the frozen cells to liquid nitrogen (vapor phase) for long-term storage.

InnoCellular Tech Pte Ltd
Block 79 Ayer Rajah Crescent, #05-03, Singapore 139955
(E) enquiry@Innocellular.com | www.innocellular.com.sg

MesenPlify™ sXF

Product Sheet

Catalogue Number C0001

Characterization of hMSC

hMSCs cultured can be characterized with flow cytometry, colony forming unit assay, and multi-lineage differentiation assay.

Adaptation of hMSC to MesenPlify™ sXF media

In case of using other brands for hMSCs culture, hMSCs can be directly adapted to complete MesenPlify™ sXF media without prior conditioning.

Warranties

For Research Use Only (RUO), or for further manufacturing.
Not for Human or Animal Diagnostic or Therapeutic use.

Disclaimer: While InnoCellular Tech has taken all reasonable measure to ensure the accuracy and correctness of the information provided, it does not provide any warranties concerning the accuracy of such information. InnoCellular warrants its products meet the applicable standards as documented in the COA when used in accordance with the recommendations in the product sheet and within the stated shelf life of the product.

In the event of a breach of the warranty, InnoCellular Tech's sole obligation shall be to replace, at its discretion, the relevant product or part thereof, upon prompt notification from the customer. In instances where reasonable efforts to repair or replace the product prove unsuccessful, InnoCellular Tech will provide a full refund of the amount paid for the product.

It is important to note that InnoCellular Tech shall not be liable for any consequential, incidential, special, or any other damages resulting from economic loss, or property damage experienced by any customer due to the use of InnoCellular Tech's product.