

OPM-LMH SFM1

Serum-free Cell Culture Medium

— For Vaccine Production

OPM-LMH SFM1 is a serum-free cell culture medium designed for suspension culture of Imh cells and contains L-Glutamine. This medium supports the production of fowl adenovirus vaccine.

Application

OPM-LMH SFM1 is intended for large scale manufacturing of therapeutic biomolecules, as well as for research purposes, but not for human or any therapeutic use.

Storage & Transportation

Store at 2~8°C, dark and dry
Ship at Room temperature (Liquid), Blue ice (Dry powder)

Shelf Life

OPM-LMH SFM1 Dry Powder: 24 months
OPM-LMH SFM1 Medium Liquid: 12 months

Reconstitution Method for Dry Powder

1. Measure out 90% of final required volume of purified water intended for cell culture use, e.g. WFI. Recommended water temperature is 25~35°C (minimum final volume \geq 1L).
2. Slowly add dry powder medium at 20.21 g/L and stir for 20 minutes.
3. Add 2.22 g/L NaHCO₃ to the solution and continue to stir for 20 minutes.
4. Adjust pH to 8.5 with 5N NaOH and continue to stir until completely dissolved.
5. Adjust pH to 7.0 with 5N HCl.
6. Add cell culture grade purified water to 100% final volume.
7. Continue to stir for 10 minutes. Sterile filter using a membrane filter with a pore size of 0.22 micron.

Quality Specifications

Specifications	OPM-LMH SFM1 DPM
Appearance	Khaki powder
pH	7.0~7.5
Osmolality (mOsm/kg)	250~300
Solubility	Good by following the reconstitution instructions
Endotoxin (EU/mL)	<2.0

Cell Culture Conditions

37°C, 80% humidity, 5~8%CO₂
Shaker speed 110-150 rpm (amplitude: 50mm).

Cell Recovery

1. Rapidly thaw (<2 min) a vial of frozen cells in a 37 °C water bath.
2. Transfer the entire contents aseptically into a 125 mL shake flask containing 30 mL prewarmed OPM-LMH SFM1 cell culture medium.
3. Incubate at 37 °C in a humidified atmosphere of 5%~8% CO₂ in air on a shaker (rotating at 110~130 rpm (amplitude: 50mm)).
4. Passage the cells for at least twice until fully recovery. Proceed according normal procedure after the Population Doubling Time stays stable.

Cell Culture Passaging

1. Prewarm OPM-LMH SFM1 cell culture medium at 37 °C for 20~30min.
2. Proceed if VCD $\geq 1 \times 10^6$ /mL & viability $\geq 90\%$. Cultures should be passaged during the mid-log phase.
3. Determine the correct volume of cell culture to inoculate a new flask at a starting cell density of 0.5×10^6 cells/mL in prewarmed OPM-LMH SFM1 cell culture medium
4. Incubate flasks in a humidified 37 °C incubator with 5%~8% CO₂ on an orbital shaker at 110~150rpm (amplitude: 50mm).
5. Passage cells by repeating the above steps every 2~3 days.

Medium Adaptation

Direct Medium Adaptation

1. Cell lines may be adapted directly from serum-free media into OPM-LMH SFM1 cell culture medium. The seeding cell density can be referred to the passaging instructions or should be determined individually.
2. Cells should be passaged for a few times.
3. Adaptation is completed when the cultures attain stable VCD of 2×10^6 /mL and viability $\geq 90\%$ within 3~4 days over at least 2~3 passages.

Sequential Medium Adaptation

1. For certain cell lines cultured in serum-free media, or in presence of 5~10% serum, sequential adaptation method is recommended.
2. Monitor the cell growth until the cell density has reached $\geq 2 \times 10^6$ cells/mL.
3. Dilute the cells with a ratio of 25:75 (OPM-LMH SFM1 vs current medium), and then further dilute the culture until the cells grow well under this condition. Increase the proportion of OPM-LMH SFM1 in each subsequent operation, as is shown in the table.
4. Adaptation is completed when the cultures in 100% OPM-LMH SFM1 Cell culture medium attain stable VCD of 2×10^6 /mL and viability $\geq 85\%$ within 3~4 days over at least 2~3 passages.

OPM-LMH SFM1: current medium (%)	Seeding density ($\times 10^5$ cells/mL)	Evaluation of cell growth	Acceptance criteria for next step
25 : 75	3 ~ 4	VCD & Viability	VCD $\geq 2 \times 10^6$ /mL, Viability $\geq 90\%$ over 2 passages
50 : 50	3 ~ 4	VCD & Viability	VCD $\geq 2 \times 10^6$ /mL, Viability $\geq 90\%$ over 2 passages
75 : 25	3 ~ 4	VCD & Viability	VCD $\geq 2 \times 10^6$ /mL, Viability $\geq 90\%$ over 2 passages
90 : 10	3 ~ 4	VCD & Viability	VCD $\geq 2 \times 10^6$ /mL, Viability $\geq 90\%$ over 2 passages
100 : 0	3 ~ 4	VCD & Viability	VCD $\geq 2 \times 10^6$ /mL, Viability $\geq 90\%$ over 2 passages

Cryopreservation

1. Harvest the desired quantity of cells in mid-log phase of growth with viability over 90%.
2. Determine VCD to ensure that the final cell density is $> 1 \times 10^7$ /ml.
3. Prepare the freezing medium consisting of 90% OPM-LMH SFM1 Cell culture medium and 10% dimethyl sulfoxide (DMSO). Let the freezing medium cool down to 4°C.
4. Harvest cells by centrifugation at 400xg for 5 minutes. Remove the supernatant and resuspend the cell pellet with the cold freezing medium at $> 1 \times 10^7$ /ml.
5. Transfer the suspension to sterile cryo-vials.
6. Place the vials in a cryo-box or a controlled rate freezing apparatus following standard procedures (1°C decrease per minute).
7. For long-term storage, transfer the vials to liquid nitrogen.

Order Information

Cell Culture Media

Name	Cat No.	Type	Volume
OPM-LMH SFM1 DPM	C679018	Dry powder	50L/100L/500L
OPM-LMH SFM1 Medium	C221012	Liquid	1000mL