Optimization
Makes Differences

OPM-ST SFM3

Serum-free Cell Culture Medium

—— For Vaccine Production

OPM-ST SFM3 is a serum-free cell culture medium designed for suspension culture of swine testis (ST) cells and contains L-Glutamine. This medium supports the production of vaccines such as swine fever vaccine, swine transmissible gastroenteritis virus vaccine, porcine epidemic diarrhea vaccine, porcine parvovirus vaccine and pseudorabies virus vaccine, etc.

Application

OPM-ST SFM3 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℃, dark and dry
Ship at Room temperature (Liquid), Blue ice (Dry powder)

Shelf Life

OPM-ST SFM3 Medium Liquid: 6 months
OPM-ST SFM3 Dry Powder: 18 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℃ (minimum final volume ≧ 1L).

2. Slowly add dry powder medium at 18.65 g/L and stir for 20 minutes.

3. Add 2.3 g/L NaHCO3 to the solution and continue to stir for 10 minutes.

4. Adjust pH to 7.2 with 1N NaOH or 1N HCl.

5. Add cell culture grade purified water to 100% final volume.

6. Continue to stir for 10 minutes. Sterile filter using a membrane filter with a pore size of 0.22 micron.

Cell Culture Conditions

37℃，5~8%CO_2

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-ST SFM3 cell culture medium.
3. Incubate at 37 °C in a humidified atmosphere of 5%~8% CO_2 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-ST SFM3 cell culture medium at 37 ℃ for 20~30min.
2. Proceed if VCD ≥1×10^6/mL & viability ≥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-ST SFM3 cell culture medium
4. Incubate flasks in a humidified 37 °C incubator with 5%~8% CO_2 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-ST SFM3 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 ≥ 90% within 3~4 days over at least 2~3passages.

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 ≥2×10^6cells/mL.
3. Dilute the cells with a ratio of 25:75 (OPM-ST SFM3 vs current medium), and then further dilute the culture until the cells grow well under this condition. Increase the proportion of OPM-ST SFM3 in each subsequent operation, as is shown in the table.
4. Adaptation is completed when the cultures in 100% OPM-ST SFM3 Cell culture medium attain stable VCD of 2×10^6/mL and viability ≥ 85% within 3~4 days over at least 2~3 passages.

OPM-ST SFM3: current medium (%)	Seeding density ($\times10^5$ cells/mL)	Evaluation of cell growth	Acceptance criteria for next step
25 : 75	3 ~ 4	VCD & Viability	VCD≥2×10^6/mL, Viability≥90% over 2 passages
50 : 50	3 ~ 4	VCD & Viability	VCD≥2×10^6/mL, Viability≥90% over 2 passages
75 : 25	3 ~ 4	VCD & Viability	VCD≥2×10^6/mL, Viability≥90% over 2 passages
90 : 10	3 ~ 4	VCD & Viability	VCD≥2×10^6/mL, Viability≥90% over 2 passages
100 : 0	3 ~ 4	VCD & Viability	VCD≥2×10^6/mL, Viability≥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 > 1×10^7/ml.
3. Prepare the freezing medium consisting of 90% OPM-ST SFM3 Cell culture medium and 10% dimethyl sulfoxide (DMSO). Let the freezing medium cool down to 4℃.
4. Harvest cells by centrifugation at 400xg for 5 minutes. Remove the supernatant and resuspend the cell pellet with the cold freezing medium at > 1×10^7/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℃ 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-ST SFM3 Medium	V005103-001	Liquid	1000ml
OPM-ST SFM3 DPM	V005203-010	Dry powder	10L

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