

HybriSFM-P1B

Serum-free Cell Culture Medium

— For Biomanufacturing

HybriSFM-P1B is a serum-free cell culture medium with low concentration of protein, designed for the growth of hybridoma cells. HybriSFM-P1B supports high-density suspension culture and protein antibody expression of different hybridoma cells.

Application

HybriSFM-P1B 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

HybriSFM-P1B Medium Liquid: 6 months
HybriSFM-P1B Dry Powder: 12 months

Reconstitution Method for Dry Powder

1. Take a clean preparation container. It is recommended that the one-time preparation volume should be not less than 1L.
2. Take 90% of the final prepared volume of ultra-pure water or WFI, the temperature is 25°C to 35°C.
3. Add 16.61g/L dry powder medium slowly to water. Keep stirring.
4. Add 2.45g/L Sodium Bicarbonate to the vessel. Keep stirring for 20 minutes.
5. Add 5N NaOH slowly to adjust the pH until the solution is clear.
6. Adjust pH to 7.0 with 5N HCl slowly.
7. Adjust to the final volume with ultra-pure water or WFI. Then adjust the osmolality to 320±10mOsm/kg with calculated amount of NaCl. $NaCl \text{ powder } W(g) = V_T \times (320 - MV_{Osm}) / 31.5$, V_T : Target volume, MV_{Osm} : measured value of Osm.
8. Stirring for an additional 10 minutes. Sterilize immediately by membrane filtration.
9. Store the reconstitution media at 2°C to 8°C and dark.

Quality Specifications

Specifications	HybriSFM-P1B Medium	HybriSFM-P1B DPM
pH	7.0~7.5	7.0~7.5
Osmolality (mOsm/kg)	300~340	300~340
Solubility	---	Good by following the reconstitution instructions
Endotoxin (EU/mL)	<1.0	<1.0
Sterility test	Negative	---



Cell Culture Conditions

37°C, 5~10%CO₂;

Shaker speed 125-135 rpm.

Optimization
Makes Differences

Cell Culture Passaging

Seed the hybridoma cells in a fresh medium at (0.2~0.3) x10⁶ cells/ml when the cell density reaches (2~3) x 10⁶ cells/ml and continue the culture. Dilute directly during seeding without centrifugation.

Medium Adaptation

Direct Medium Adaptation

Cell lines may be adapted directly from serum-free media into HybriSFM-P1B medium.

Sequential Medium Adaptation

1. For certain cell lines cultured in presence of serum, sequential adaptation method is recommended.
2. Start the cell adaptation when it is in low passage and log phase.
3. Serum reduction can be performed in the concentration of 10%, 5%, 2.5%, 1%, and 0%. Passage cells at each serum concentration until the cells grow well. Proceed to the next serum concentration.
4. Adaptation is completed when the cells grow well in HybriSFM-P1B medium.

Antibody Production in Fed-batch Culture

The optimal fed-batch cultures should be optimized case by case and may need to be established by DOE method. The following conditions are only for reference:

1. Seed the cells in a fresh medium at (0.3~0.6) x10⁶ cells/ml when the cell density reaches ~3 x 10⁶ cells/ml;
2. Daily sampling for cell density and biochemical analysis is recommended. Feeding can be started on the second or third day after seeding.
3. Glucose should be added to a final concentration of 6 g/L when it is lower than 3 g/L.

Recommended fed-batch feeding strategy

Method	Basal Media	High Performance Feed	Feeding strategy	Highly Concentrated Feed	Feeding Strategy
1	HybriSFM-P1B	OPM-CHO PFF05	Add 2.5% OPM-CHO PFF05 every day from D3.	CDFS36	Add 0.25% CDFS36 every day from D3
2	HybriSFM-P1B	OPM-CHO PFF05	Add OPM-CHO PFF05 every other day from D3: D3/5/7/9, 3%/5%/5%/4% of initial culture volume.	CDFS36	Add CDFS36 every other day from D3: D3/5/7/9, 0.3%/0.5%/0.5%/0.4% of initial culture volume.

Order Information

Cell Culture Media

Name	Cat No.	Type	Volume
HybriSFM-P1B	H081801-001	Liquid	1000ml
HybriSFM-P1B DPM	H091801-050	Dry powder	50L
	H091801-010	Dry powder	10L

High Performance Feed

Name	Cat No.	Type	Volume
OPM-CHO PFF05	F81279-001	Liquid	1000ml
OPM-CHO PFF05 DPM	F91279-010	Dry powder	10L

Highly Concentrated Feed

Name	Cat No.	Type	Volume
CDFS36	C217836	Liquid	500ml/1000ml
CDFS36 DPM	C672069	Dry powder	1L/2L/5L/10L/50L/100L

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