Optimization
Makes Differences

OPM-CHO CD08

Chemically Defined Cell Culture Medium for Transfection

—— For Biomanufacturing

OPM-CHO CD08 is an animal origin-free, chemically defined cell culture medium that contains no proteins, growth factors, hydrolysates, or components of unknown composition and is developed for the growth of Chinese Hamster Ovary (CHO) cells and transfection in suspension culture. The medium is formulated without hypoxanthine and thymidine for use in dihydrofolate reductase (DHFR)-amplified systems, without L-glutamine for use in glutamine synthetase systems, and without phenol red to minimize estrogen-like effects of phenol red.

Application

OPM-CHO CD08 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-CHO CD08 Medium Liquid: 12 months
OPM-CHO CD08 Dry Powder: 24 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 16.05 g/L while stirring, and continue mixing for 10 minutes. Residual powder attached to the vessel wall should be taken into the solution.

3. Add 2.22 g/L NaHCO3 to the solution, and continue to stir.

4. Adjust pH to 8.3~8.5 with 5N NaOH and stir for 20~30 minutes until completely dissolved.

5. Adjust pH to 7.0 by slowly adding 5N HCl.

6. Add cell culture grade purified water to 100% final volume. Continue to stir for 5 minutes. Adjust pH to 7.0 using 5N HCl or 5N NaOH.

7. Adjust osmolality to 285±10 mOsm/kg using NaCl (Calculate osmolality： required NaCl(g)= final solution volume(L)×(285 – measured value) / 31.5).

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

Quality Specifications

Specifications	OPM-CHO CD08 Medium	OPM-CHO CD08 DPM
Appearance	Red clear liquid	Off -white or light yellow powder
pH	7.0~7.4	7.0~7.4
Osmolality (mOsm/kg)	270~300	270~300
Solubility	——	Good by following the reconstitution instructions
Endotoxin (EU/mL)	＜1.0	＜1.0
Sterility test	Negative	——

Cell Culture Conditions

37 °C, 80% humidity, 5~8%CO_2

Shaker speed 110–150 rpm (amplitude: 50mm).

Cell Recovery

1. Incubate the original medium and OPM-CHO CD08 medium at 37 °C, and add 6 mM L-glutamine before use.

2. Recover cells according to the original medium method.

Cell Culture Passaging

1. Incubate the original medium and OPM-CHO CD08 medium at 37 °C, and add 6 mM L-glutamine before use.

2. Passage cells every 2~3 days to keep cells in the early-log phase.

3. Seed cells at a density of (0.3~0.6)×10^6 cells/ml.

4. Cells should be passaged if VCD = (3~4)×10^6/mL & viability ≥95% (2-4 days).

Note: It is critical to keep the seed cells in the log phase, and different types of CHO cells may have different log phase ranges.

5. Repeat the above steps to preserve cells or expand cells for transfection and expression.

Medium Adaptation

It is critical that cell viability be ≥95% and the growth rate be in mid-logarithmic phase prior to initiating adaptation procedures.

Direct Medium adaptation

In most cases, CHO cells in other media can directly adapt to OPM-CHO CD08 medium following the previous daily subculture protocol.

Sequential Medium adaptation

If suboptimal performance is achieved using the direct adaptation method, use the sequential adaptation method. Incubate the original medium and OPM-CHO CD08 medium at 37 °C, and add 6 mM L-glutamine before use. Some key points are recommended as followings,

1) Select the cells at lower generation and ensure the cells in the logarithmic phase.

2) Resuscitate cells using the original medium, and continue to use the original medium to subculture 2-3 generations to achieve stable cell growth.

3) When the cell density reached 3~4 x10^6 cells/ml, the cells were inoculated with 0.6 x10^6 cells/ml into the medium containing 1/3 volume of OPM-CHO CD08 and 2/3 volume of original medium.

4) When the cell density reached 3x10^6 cells/ml and the cell viability is greater than 95% (3-4 days), the cells were inoculated with 0.5x10^6 cells/ml into the medium containing 2/3 volume of OPM-CHO CD08 and 1/3 volume of original medium.

5) When the cell density reached 3x10^6 cells/ml and the cell viability was greater than 95% (3-4 days), the cells were inoculated into 100% OPM-CHO CD08 medium with 0.4x10^6 cells/ml.

6) The cells were inoculated in OPM-CHO CD08 medium at a density of 0.3x10^6 cells/ml, and continue to subculture 2-3 generations to achieve stable cell growth.

Recommended transfection conditions

Transfection conditions need to be optimized according to specific cell line/protein molecule. The following conditions are only for reference,

VCD	(5~ 6)x10^6 cells/ml, cell viability> 95%
DNA	(~1)mg/L
PEI	5~8mg / L

Recommended feeding strategy

Protein production may be enhanced by adding feed OPM-CHO ProFeed.

The recommended feeding strategy is as below,

Basal medium	Cell recovery after transfection	Feed strategy	
OPM-CHO CD08	Cells recover well (Doubling time of cells after transfection is not changed significantly, and the viability is greater than 90%)	OPM-CHO ProFeed	Add 4%, 5%, 6%, 5%, 4% and 4% of the initial culture volume of OPM-CHO ProFeed , at D1, D3, D5, D7, D9 and D11 respectively after transfection; When glucose is ≤ 3g / L, add glucose concentrate at 6g/L final concentration
	Cells recover not well (Doubling time of cells after transfection is prolonged significantly, or the viability is lower than 90%	OPM-CHO ProFeed	Observe and determine the feed starting point according to cell recovery

Order Information

Cell Culture Media

Name	Cat No.	Type	Volume
OPM-CHO CD08 Medium	P081308-001	Liquid	1000ml
OPM-CHO CD08 DPM	P091308-010	Dry powder	10L
	P091308-050	Dry powder	50L

Shanghai OPM Biosciences Co., Ltd.

OPM Headquarter: Building #28, 908 Ziping Road, Pudong, Shanghai, China
CDMO Development Center: Building #3, 100 Banxia Road, Pudong, Shanghai, China
Media & CDMO Manufacturing Center: C3&D3, No. 356, Zhengbo Road, Fengxian, China

+86 021-6818 2622
service@opmbiosciences.com
www.opmbio.com