

# CDM4CHO

## HYCLONE MEDIA AND SUPPLEMENTS

HyClone™ CDM4CHO is a protein-free and animal-derived component-free (ADCF) cell culture medium. This regulatory-friendly medium is developed through the HyClone Metabolic Pathway Design process (see box) to increase process yields for the industrial manufacture of recombinant proteins using a variety of CHO cell clones. CDM4CHO medium has been successfully tested in a variety of culture systems, including T-flasks, shaker flasks, and bioreactors including fed-batch and perfusion culturing. CDM4CHO medium is available in liquid and powder formats in user-friendly packaging (Fig 1).

Key features of CDM4CHO medium include

- Chemically defined and animal-derived component-free formulation
- Designed for high cell yield and recombinant protein production
- Allows for direct or sequential adaptation
- Designed for large-scale culture applications, including perfusion and fed-batch strategies
- Manufactured from traceable components and according to cGMP guidelines

## Specifications

CDM4CHO medium is a protein-free and animal-derived component-free and does not contain phenol red. CDM4CHO liquid medium contains poloxamer 188 and is available with 4 mM L-glutamine or without L-glutamine to support the glutamine synthetase (GS) gene expression system. CDM4CHO powder medium does not contain poloxamer 188 or L-glutamine.

### **Product handling**

Store medium at 2°C to 8°C, away from light. In addition, powder medium should be stored protected from moisture in a tightly sealed container.



**Fig 1.** CDM4CHO medium is available as liquid or powder in pack sizes suitable for small-volume cell culture as well as large-scale bioprocessing applications.

### **Metabolic Pathway Design process**

An optimal cell culture process is dependent on a variety of factors, such as cell line, specific clones, media, and feeds, as well as processes to maximize viable cell densities and productivity. Our experts in medium design and development know and understand these factors at the metabolic level. They evaluate each metabolic profile, understanding nutritional demands and waste creation, to make sure the correct nutrient type and quantity is used to minimize waste and resultant cell toxicity. Our experts use their understanding of metabolic pathways to optimize media for enhanced viable cell densities and productivity. Once the medium has been optimized using this Metabolic Pathway Design process, our scientists can help you devise an effective cell culture strategy using a combination of media and feeds to further enrich productivity and reduce process inefficiencies.

## Suggested preparation

### Reconstitution of CDM4CHO powder medium

1. While stirring, add CDM4CHO dry powder to cell culture-grade water at 90% of final preparation volume (17.14 g/L). Mix until dissolved. If your water source is normally cool, it might be useful to adjust the water temperature. Using warmer room temperature water (22°C to 25°C) will improve dissolution time. Mix for 20 min or until dissolved.
2. Add 1.0 g/L poloxamer 188 and 2.2 g/L sodium bicarbonate (Table 1). Mix until dissolved.
3. Bring vessel to final volume with cell culture-grade water. Allow solution to mix for 10 to 20 min.
4. Check pH and osmolality. Expected values:
  - pH 7.0 to 7.4
  - Osmolality 280 to 320 mOsm/kg
5. Sterile filter into desired container using a 0.2 µm sterile filter.

### Preparation notes

CDM4CHO powder medium does not contain L-glutamine. Recommended concentration: 4 mM.

**Table 1.** Sodium bicarbonate supplementation guide

CO <sub>2</sub> environment	Sodium bicarbonate level
Online pH control	0.25 g/L
5% CO <sub>2</sub> incubator	2.20 g/L
10% CO <sub>2</sub> incubator	3.60 g/L

**Note:** Additional buffering can be achieved by adding 5–25 mM HEPES.

## General culture recommendations

1. Cultures should be incubated at 37°C and in a 5% CO<sub>2</sub> environment.
2. Maintain adapted cells by establishing a mid-logarithmic growth phase subculturing schedule.
3. Suggested seeding density of cultures: 2.0 × 10<sup>5</sup> cells/mL; viability should be > 90%.

### Direct adaptation

1. Transfer cells grown in current medium directly into CDM4CHO at 2.0 × 10<sup>5</sup> cells/mL.
2. When viable cell density reaches 1.0 to 1.5 × 10<sup>6</sup> cells/mL, subculture the cells.
3. Cells should be subcultured every 48 to 96 h.
4. If cell viability drops below 80%, proceed to sequential adaptation.

### Sequential adaptation

Dilute serum-containing medium with an equal volume of CDM4CHO. This preparation will be referred to as the sequential adaptation medium (SAM). Prepare twice the volume of medium needed for the culture vessel in use (i.e., for a T-75 flask using 25 mL of medium, prepare 50 mL of SAM). Prior to each subculture, warm medium to 37°C.

1. Subculture the cells into SAM at a seeding concentration of 2.0 × 10<sup>5</sup> cells/mL. For best results, the culture should be ~ 70% confluent.
2. When the cells reach a density of 1.0 to 1.5 × 10<sup>6</sup> cells/mL, subculture into an equal mixture of SAM and fresh CDM4CHO at a seeding density of 2.0 × 10<sup>5</sup> cells/mL.

### Cryopreservation

CDM4CHO adapted cells can be cryopreserved in a medium consisting of a 1:1 ratio of fresh and conditioned CDM4CHO. To this add DMSO at a final concentration of 7.5%.

## Quality control testing

Quality control test specifications are listed in Table 2.

**Table 2.** Test specifications<sup>1</sup>

Appearance	Clear solution
Osmolality	280 to 320 mOsm/kg
pH	7.0 to 7.4
Sterility	No growth (bacteria or fungi)
Endotoxin	≤ 10.0 EU/mL <sup>1</sup>
Application	Growth promotion <sup>1</sup>

<sup>1</sup> Refer to certificate of analysis for actual results.

## Custom production

Formulations and delivery systems can be customized to your specific process requirements or optimized to maximize process yields.

### Rapid Response Production (RRP)

Our RRP program manufactures up to 200 L of your custom prototype formulation within seven working days of your request. Use our RRP service to expedite the development and testing of custom media for your biopharmaceutical manufacturing process.

**Table 3.** Supplement matrix

	Amino acids	Vitamins	Glucose	Trace elements	Growth factors	Hypoxanthine/ thymidine	ADCF* lipids	ADCF* cholesterol	Suitable for	Product code
Cell Boost™ 1 Supplement (R05.2)	•	•	•						HEK293 CHO	SH30584
Cell Boost 2 Supplement (R15.4)	•		•						PER.C6™ CHO	SH30596
Cell Boost 3 Supplement (JM3.5)	•	•	•	•		•			Hybridoma Myeloma	SH30825
Cell Boost 4 Supplement (PS307)	•	•	•	•	•		•	•	CHO	SH30857
Cell Boost 5 Supplement (CN-F)	•	•	•	•	•	•	•	•	Hybridoma NS0 HEK293 CHO	SH30865
Cell Boost 6 Supplement (CN-T)	•	•	•	•	•	•	•	•	T-Cells Hybridoma NS0 HEK293 CHO	SH30866
LS250 supplement							•	•	NS0	SH30554
LS1000 supplement								•	NS0	SH30555

\* Animal-derived component-free

## Related products

Table 3 gives an overview of HyClone supplements.

### **HyClone Cell Boost kit**

Cell Boost Process Supplements (100 g each) contain samples of supplements designed to increase cell productivity in a variety of cell lines. Each supplement is developed through the Metabolic Pathway Design process and is chemically-defined and protein-free with no animal derived components.

### **HyClone LS250 supplement**

LS250 is a chemically defined, ADCF lipid supplement developed to stimulate cell growth and monoclonal antibody (MAb) production in NS0 cell cultures using traditional hybridoma serum-free media.

### **HyClone LS1000 supplement**

LS1000 supplement is a chemically defined, ADCF lipid supplement developed to stimulate cell growth and MAb production in NS0 cell cultures using traditional hybridoma serum-free media.

The supplement is formulated using a proprietary complexing process for enhanced cholesterol delivery. LS1000 has been successfully tested in a variety of serum-free medium cultures, including HyClone CDM4NS0 medium and HyClone CDM4MAb medium.

# Ordering information

CDM4CHO medium is manufactured in homogenous liquid lot sizes up to 10 000 L and powder lots up to 250 000 L.

<b>Product</b>	<b>Size</b>	<b>Product code</b>
HyClone CDM4CHO liquid medium With L-glutamine	1000 mL bottle	SH30557.02
	6 × 1000 mL bottles	SH30557.LS
	5 L bag	SH30557.03
	10 L bag	SH30557.04
	20 L bag	SH30557.05
	50 L bag	SH30557.06
	100 L bag	SH30557.07
	200 L bag	SH30557.08
	500 L bag	SH30557.09
HyClone CDM4CHO liquid medium Without L-glutamine	1000 mL bottle	SH30558.02
	6 × 1000 mL bottles	SH30558.LS
	5 L bag	SH30558.03
	10 L bag	SH30558.04
	20 L bag	SH30558.05
	50 L bag	SH30558.06
	100 L bag	SH30558.07
	200 L bag	SH30558.08
	500 L bag	SH30558.09
HyClone CDM4CHO powder medium Without L-glutamine	1 × 5 L	SH30556.01
	1 × 10 L	SH30556.02
	1 × 50 L	SH30556.03
	1 × 100 L	SH30556.04
	1 × 500 L	SH30556.05
	1 × 1000 L	SH30556.06

<b>Related products</b>	<b>Size</b>	<b>Product code</b>
HyClone Cell Boost kit	6 × 100 g	SH30890
HyClone LS1000 cholesterol supplement	50 mL bottle	SH30554.01
	100 mL bottle	SH30554.02
	500 mL bottle	SH30554.03
	1000 mL bottle	SH30554.04
HyClone LS250 lipid supplement	100 mL bottle	SH30555.01
	500 mL bottle	SH30555.02
	1000 mL bottle	SH30555.03

## [cytiva.com/hyclone](https://www.cytiva.com/hyclone)

Cytiva and the Drop logo are trademarks of Global Life Sciences IP Holdco LLC or an affiliate. Cell Boost and HyClone are trademarks of Global Life Sciences Solutions USA LLC or an affiliate doing business as Cytiva.

PER.C6 is a trademark of Crucell. All other third-party trademarks are the property of their respective owners.

© 2020 Cytiva

All goods and services are sold subject to the terms and conditions of sale of the supplying company operating within the Cytiva business. A copy of those terms and conditions is available on request. Contact your local Cytiva representative for the most current information.

For local office contact information, visit [cytiva.com/contact](https://www.cytiva.com/contact)

CY12725-17Apr20-DF

