HyClone™ media and supplements

CDM4PERMAb

HyClone CDM4PERMAb medium has a chemically defined and animal-derived component-free (ADCF) formulation. This high-performance cell culture medium has been developed through the Metabolic Pathway Design process (see box) to increase process yields in the production of human antibodies and recombinant proteins using PER.C6™ technology. The medium has been successfully tested in a variety of applications, including fed-batch bioreactor cultures. CDM4PERMAb is available in liquid and powder formats in user-friendly packaging (Fig 1).

Key features of CDM4PERMAb include

- Animal-derived component-free
- Chemically defined formulation
- Designed for high cell yield and recombinant protein production
- Allows for direct or sequential adaptation
- Component traceability
- Manufactured according to cGMP guidelines

Specifications

- Does not contain L-glutamine
- Does not contain phenol red
- Liquid medium contains poloxamer 188
- Store medium at 2°C to 8°C, away from light.
- Powder medium should be stored protected from moister in a tightly sealed container.

Suggested preparation

Reconstitution of CDM4PERMAb powder medium

1. While stirring, add CDM4PERMAb powder medium to cell culture-grade water at 90% of final preparation volume (17.3 g/L). If your water source is normally cool, it may be useful to adjust the water temperature. Using warmer room temperature water (22°C to 25°C) will improve solubilization time. Mix for 20 min or until dissolved. Medium should be a clear, golden solution at this point.

Fig 1. CDM4PERMAb medium is available as liquid or powder in pack sizes suitable for small-volume cell culture as well as large-scale bioprocessing applications.

2. Add 0.5 g/L of poloxamer 188 and 3.2 g/L of sodium bicarbonate. Ensure each component has completely dissolved before adding the next component.
3. Bring vessel to final volume with culture grade water. Allow solution to mix for 10 to 20 min.

Metabolic Pathway Design process

An optimal cell culture process is dependent of a variety of factors including the parental cell line, the genetic makeup of the specific clone, medium and feed composition, as well as process variables to maximize viable cell densities and titers while maintaining cell morphology. Our experts in medium design and development know and understand how these factors can influence the metabolic processes involved. They evaluate the culture's metabolic activities, measuring nutritional demand and waste creation to make sure the correct type and quantity of nutrients are used to minimize waste and resultant cell toxicity. Our experts use their understanding of metabolic pathways to optimize medium composition for enhanced productivity and viable cell densities. Once a medium has been optimized using this Metabolic Pathway Design process, our scientists can help you devise the most effective cell culture strategy using a combination of medium and feeds to further enrich productivity and reduce process inefficiencies.

4. Adjust pH to between 7.0 and 7.2 by adding 1 N NaOH, or 1 N HCl drop wise to solution.
5. Check osmolality. Expected value is 290 to 340 mOsm/kg.
6. Sterile filter into desired container using a 0.2 µm sterile filter.

Preparation notes

CDM4PERMAb powder medium does not contain L-glutamine. Recommended concentration: 4 mM.

General culture recommendations

1. Cultures should be incubated at 37°C in a 5% CO_2 environment.
2. Maintain stock cells in classical medium with serum supplementation.
3. Upon subsequent passage, following trypsinization, seed two new T-flasks. One flask will be maintained at the current serum concentration as a backup, while the other will be reduced to half that serum concentration. This serum depletion process is repeated until reaching a concentration of 2.5% fetal bovine serum (FBS).
4. Re-plate cells at 2.5% FBS and incubate for 24 h to allow attachment and spreading.
5. Following the 24 h incubation, replace the serum-containing medium with CDM4PERMAb medium. Cells will not attach firmly and should be passaged without trypsin.
6. A passage schedule of 3 to 4 days should be maintained for 2 to 3 passages. Adaptation is complete once cells have transitioned to a doubling rate of 24 h or less per doubling.

Cell maintenance

Maintain adapted cells by establishing a passage schedule that allows the cells to be passed while in log growth phase. PER.C6 cells cultivated in CDM4PERMAb medium for routine maintenance should be subcultured every 72 to 96 h. The passage schedule and seeding density may be adjusted to optimize performance. The recommended population seeding density of new cultures for general maintenance is 0.3×10^6 cells/mL. The culture viability of an adapted culture should remain greater than 90%. During adaptation from serum-containing medium, however, viabilities might be slightly lower than 90%. Cells should exhibit a population doubling time of approximately 24 to 30 h. If the recommended population seed density of 0.3×10^6 cells/mL is used, cultures typically reach peak cell population densities of between 8.0 and 12.0×10^6 cells/mL, depending on the specific clone used. Doubling times during an adaptation period might be higher.

Cryopreservation

CDM4PERMAb medium adapted cells can be cryopreserved in a medium consisting of a 1:1 ratio of fresh and conditioned CDM4PERMAb medium. To this, add DMSO to a final concentration of 7.5%.

Quality control testing

Quality control test specifications are listed in Table 1.

Table 1. Test specifications[1]

Appearance	Clear yellow solution
Osmolality	290 to 340 mOsm/kg
pH	7.0 to 7.4
Sterility	No growth (bacteria or fungi)
Endotoxin	≤ 1 EU/mL[1]
Application	Growth promotion

[1] 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 buffers and process liquids for your biopharmaceutical manufacturing process.

Related products

Table 2 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 (Table 2). 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, animal-derived component-free 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, animal-derived component-free 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 and CDM4MAb media.

Table 2. Supplement matrix

	Amino acids	Vitamins	Glucose	Trace elements	Growth factors	Hypoxanthine/ thymidine	ADCF* lipids	ADCF* cholesterol	Suitable for	Code number
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

Ordering information

CDM4PERMAb is manufactured in homogenous liquid lot sizes up to 10 000 L and powder lots up to 250 000 L.

Product	Size	Code number
HyClone CDM4PERMAb liquid medium	500 mL bottle	SH30871.01
	1000 mL bottle	SH30871.02
	10 L bag	SH30871.05
	20 L bag	SH30871.06
	50 L bag	SH30871.07
	100 L bag	SH30871.08
	200 L bag	SH30871.09
	500 L bag	SH30871.10
	900 L bag	SH30871.11
HyClone CDM4PERMAb powder medium	1 × 5 L*	SH30872.01
	1 × 10 L*	SH30872.02
	1 × 50 L*	SH30872.03
	1 × 100 L*	SH30872.04
	1 × 500 L†	SH30872.05
	1 × 1000 L†	SH30872.06

* High-density polyethylene (HDPE) bottle

† Polybag/pail

Related products	Size	Code number
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

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