#### EIV22024E

# **ExCell**

# **ExCell Bio**

# **OptiVitro® CHO Serum-free Basal Medium CE01**

For Research and Manufacturing Use

Not Intended for Diagnostic and Therapeutic Use

## **User Manual**

Catalog Number





## **PRODUCT DESCRIPTION**

OptiVitro<sup>®</sup> CHO Serum-free Basal Medium CE01 is a state-of-the-art, animal-free, protein-free, and chemically-defined medium that is specifically designed for the high-density culture of CHO-K1, CHO-DG44, and CHO-S etc. It is an ideal medium for achieving high-level expression of recombinant proteins, while eliminating concerns over potential contamination from animal-derived components.

## SPECIFICATION, STORAGE AND TRANSPORTATION

## REQUIREMENT

Product Name	Cat.#	Specification	Storage	Transportation	Shelf Life
OptiVitro® CHO Serum-free Basal Medium CE01	CE000-N031	500 mL Liquid	2-8°C Protect From Light	< 25°C	12 months
	CE000-N032	1000 mL		Protect From	
		Liquid		Light	
	CE000-N033	1 L			
		Powder	2-8°C Dark and Dry	< 10°C	24 months
	CE000-N034	10 L			
		Powder		Protect From	
	CE000-N035	100 L		Light	
		Powder			

## **PERFORMANCE, APPLICATION AND RESTRICTION**

1. Please make sure to store the cell culture medium in a light-protected environment, avoid fluorescent lamps or other lamplight exposure, and better to use colored packaging bags in the refrigerator or warehouse.

2. During the transportation of products, try to avoid the impact of fluorescent lamps or other lamplight exposure on the appearance of the product, resulting in appearance discoloration.

3. During the transportation of the product to the clean area, it is essential to carry out a cleaning and sterilization process. The sterilization method may involve disinfectant wiping, and not utilize UV irradiation for sterilization.

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**(**Note **)** When passing through transfer windows equipped with UV sterilization, remember to proactively switch off the UV lamp inside the transfer window.

## **INSTRUCTION FOR USE**

#### **Medium preparation**

1. Measure 80% of the final volume WFI or distilled water in a clean vessel.

2. Add 22.15g/L OptiVitro<sup>®</sup> CHO Serum-free Basal Medium CE01 powder slowly to the water, mix for 30 minutes.

- 3. Adjust the pH to 8.5 with 5 N NaOH solution. After adjusting, continue stirring for an additional 10 minutes.
- 4. Adjust the pH to 7.0 with 6 N HCl solution. After adjusting, continue stirring for an additional 5 minutes.
- 5. Add 2.317g/L NaHCO $_3$  and mix for 10 minutes.
- 6. QS to final production volume and mix for 5 minutes.
- 7. Measure the final pH (6.90-7.50).
- 8. Measure the osmolality ( 270-300 mOsm/kg).
- 9. Sterilize immediately by membrane filtration (< 0.22 microns).

## Culture conditions

Suggested culture condition, Temperature:37°C, RH:80%, CO<sub>2</sub>:5%, 120-150rpm.

## Adaptation of cells

OptiVitro<sup>®</sup> CHO Serum-free Basal Medium CE01 is a highly versatile medium that can be seamlessly integrated into most stable cell lines. We recommend direct adaptation of CHO cells to OptiVitro<sup>®</sup> CHO Serum-free Basal Medium CE01, as this can often be achieved without the need for a stepwise adaptation process. In cases where direct adaptation is unsuccessful, sequential adaptation is suggested.

It's important to note that some serum-free media (SFM) may contain hydrolysates or growth factors, which can cause cells cultured in such media to become dependent on these additives. Therefore, when transitioning from SFM to a chemically-defined (CD) culture medium like OptiVitro<sup>®</sup> CHO Serum-free Basal Medium CE01, certain cell lines may require adaptation to ensure optimal growth and performance. Our technical experts are available to provide guidance on the adaptation process for your specific cell line.

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#### **Direct adaptation**

1. Recover cells: Recover cells using the original medium (15-30 mL culture volume in a 125 mL shake flask). Subculture cells until consistent growth is achieved (approximately 3 passages). It is crucial that cell viability is at least 90% throughout the process.

2. Replacement medium: Transfer cells into 100% OptiVitro<sup>®</sup> CHO Serum-free Basal Medium CE01 at a seeding density of  $0.3 \times 10^6 - 0.8 \times 10^6$  viable cells/mL. Incubate at 37°C in a humidified atmosphere of 5% CO<sub>2</sub> in air on an orbital shaker platform rotating at 120–140 rpm (30-40 mL culture volume in a 125 mL shake flask). Adjust according to your existing culture method.

#### Sequential adaptation

1. Recover cells: Recover cells using the original medium (15-30 mL culture volume in a 125 mL shake flask). Subculture cells until consistent growth is achieved (approximately 3 passages). It is crucial that cell viability is at least 90% throughout the process.

Replacement medium: At each subsequent passage, dilute cells with stepwise increasing ratios of OptiVitro<sup>®</sup>
CHO Serum-free Basal Medium CE01 and the original medium (e.g. 50:50, 75:25, 90:10, 95:5, 100:0). At least
2 passages at each step are recommended to ensure that cells appropriately adjust to the new medium.

3. Culture optimization: Monitor cell growth and viability throughout the adaptation process. Adjust the ratios of the old and new media as needed to optimize cell growth and viability.

[Note] Please note that the proportion of new and old culture medium should be adjusted appropriately based on your specific cell line and culture conditions. Sequential adaptation may require more time and effort than direct adaptation, but it can be an effective method for ensuring successful transition to OptiVitro<sup>®</sup> CHO Serum-free Basal Medium CE01. Our technical experts are available to provide guidance on the adaptation process for your specific cell line.

#### Cryopreservation

1. Prepare the desired quantity of cells, harvesting them in mid-log phase of growth with >90% viability.

2. Prepare the required volume of cryopreservation medium consisting of 90% OptiVitro<sup>®</sup> CHO Serum-free Basal Medium CE01 and 10% dimethyl sulfoxide (DMSO) freshly prepared.

3. Harvest cells by centrifugation at 1,200 rpm for 5 minutes.

4. Resuspend the pellet in the pre-determined volume of cold cryopreservation medium (suggested cell density:

 $1-2 \times 10^7$  viable cells/mL).

- 5. Freeze the cells in a controlled rate freezing apparatus following standard procedures.
- 6. For long-term storage, transfer the vials to a liquid nitrogen tank (vapor phase).

## **DISCLAIMER**

1. The product should be used according to the instructions in the manual. If the experimenter fails to operate according to the instructions, our company will not be responsible for any deviation in product performance caused by this.

2. The product is only used for scientific research and commercial production, and is not suitable for clinical diagnosis and treatment. Otherwise, all consequences arising shall be borne by the experimenter, and our company shall not be responsible.