

The User's Manual—Tobitec FedVigor02

Basic Information Introduction

Product Introduction

Tobitec FedVigor02 is chemically-defined (CD) feed media, contains amino acids, vitamins, glucose, inorganic salts and trace elements, without protein, protein hydrolysate, growth factor and any animal-derived components. Tobitec FedVigor02 is an efficient feeding formula specially designed to improve the growth and transient transfection expression of CHO and HEK293 cells, which can meet the nutritional requirements of high-density cell culture and high product expression.



Application Scope

Tobitec FedVigor02 can be applied to the transient transfection expression of CHO and HEK293 cells, effectively improving the cell density and product expression. It can be used in the research and further manufacturing of protein/antibody and virus related products, but cannot be directly used in the human body or used as a medication.

Shipping, Storage and Validity Period

Product	Catalog No.	Storage	Shipping	Validity period
Tobitec FedVigor02	LQ47, Liquid	2°C ~ 8°C, Protect from light	2°C ~ 8°C, Protect from light	6 months
Tobitec FedVigor02	DP47, Powder	2°C ~ 8°C, Protect from light	2°C ~ 8°C, Protect from light	24 months

Protocol for Hydration of Powder Medium

1. Fill the mixing container with purified water (20 ~ 30°C) at 70% of the final volume.
2. Slowly add 209.14 g/L of powder medium with gentle stirring. Mix for 30 minutes.
3. Adjust the pH to 6.8 ~ 7.2 using 5M NaOH solution. Mix for more than 30 minutes.
4. Adjust the solution volume to 100% with purified water. Mix for 5 ~ 10 minutes.
5. Filter immediately the media with a 0.22 µm membrane filter.

Quality Index of Powder and Liquid Media

Product Index	Tobitec FedVigor02 (LQ47), Liquid	Tobitec FedVigor02 (DP47), Powder
Appearance	Red, clear liquid	Off-white powder
pH	6.6 ~ 7.4	6.6 ~ 7.4 (pre-filter)
Osmolality (mOsmol/kg)	240 ~ 340 (5-fold dilutions)	240 ~ 340 (5-fold dilutions)
Solubility	--	Dissolve well according to the protocol for hydration of powder medium
Endotoxin (EU/mL)	< 10	< 10
Sterility	Negative	--
Bioburden	--	Aerobic bacteria: < 200CFU/g Molds and yeasts: < 50CFU/g

Reference Fed Feeding Strategy

Application of Tobitec FedVigor02 in CHO Cell Transient Expression

- CHO cells that have completed adaptive culture can be transfected after 3 times of subculture. Fresh medium was prepared before transfection. If the cell line was not Expi CHO, the final concentration of EDTA was 4mg/L.
- 150g ~ 300g (about 800rpm ~ 1200rpm) centrifuge for 5 min to treat transfected cells, and re-suspension with fresh medium;
- Live cell density and cell viability were measured by sampling, and fresh medium was added according to the counting results, and the live cell density was diluted to $4 \sim 6 \times 10^6$ cells/mL;
- The optimal transfection conditions should be optimized according to the specific situation when performing transfection operations. The following transfection conditions are for reference only:

VCD	DNA	PEI
$4 \sim 6 \times 10^6$ cells/mL, Via > 95%	1.5mg/L	7.5mg/L

- After CHO cell transient operation, culture cells under specified environmental conditions and fed according to the following feeding strategy:

Feeds/Time	D1	D4	D7
Tobitec FedVigor02	5 ~ 8%	5 ~ 8%	5 ~ 8%
Glucose	When the glucose concentration is below 4 g/L, glucose is added at the final concentration of 6 g/L		
Glutamine	Control glutamine 2 ~ 6 mmol/L after transfection or add 4 mmol/L glutamine according to culture volume during feeding		

Application of Tobitec FedVigor02 in HEK293 Cell Transient Expression

1. HEK293 cells that have completed adaptive culture can be transfected after 3 times of subculture. Fresh medium was prepared before transfection.
2. HEK293 cells were seeded into fresh medium at a viable cell density of 0.5×10^6 cells/mL.
3. After 2 ~ 3 days of cell culture, the viable cell density can reach $2 \sim 4 \times 10^6$ cells/mL, dilute the culture with the same volume of fresh medium (volume ratio = 1 : 1), continue to culture the cells, and take samples every day to detect the viable cell density ($\times 10^6$ cells/mL) and cell viability (%).
4. The optimal transfection conditions should be optimized according to the specific situation when performing transfection operations. The following transfection conditions are for reference only:

VCD	DNA	PEI
4.5×10^6 cells/mL, Via > 95 %	1.5mg/L	4.5mg/L

5. After HEK293 cell transient operation, culture cells under specified environmental conditions and fed according to the following feeding strategy:

Feeds/Time	D0	D1	D3
Tobitec FedVigor02	--	5 ~ 8%	5 ~ 8%
Glucose	--	4mmoL/L	mmoL/L
Glutamine	When the glucose concentration is below 3 g/L, glucose is added at the final concentration of 5 g/L		
Parameter recommendation: the initial culture temperature after transfection is 36.5°C, no cooling, pH: 7.1 ± 0.2, DO: 40%.			

【Technical Support】:

According to the terms of sales, please contact our technicians with any problems :

Tel: +86 21-64909996-393 Fax: +86 21-64909996-730

Email: bioreagent@tofflon.com

