

# CELL | ST™

Cell culture media  
**For better lives**

CELLiST™ F7 Feed Media

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# CELLiST™ F7 Feed Media

## Overview

The CELLiST™ product line provides an all-in-one solution for all your biologics manufacturing needs. CELLiST™ F7 feed medium is a single-agent, easy to use, high performance feed medium suitable for any CHO cell line in any stage of the biopharmaceutical process. CELLiST™ F7 feed medium is a completely chemically-defined, animal origin-free medium. CELLiST™ F7 feed medium was developed incorporating Ajinomoto Group's life-long experience in amino acids science and cutting edge Digital Twin technology in collaboration with A\*STAR Bioprocess Technology Institute's (BTI). CELLiST™ F7 medium utilizes Ajinomoto Group's proprietary cysteine-control technology to achieve optimal cell culture performance and productivity while keeping a single-agent feed system for user convenience. In addition, the key components of CELLiST™ F7 feed medium was optimized through the use of bio-simulations, multiomics and AI technologies, focusing on increasing performance of specific metabolic pathways, such as the TCA cycle. This integration of years of expertise with innovative technologies will help you achieve the most out of your CHO fed-batch process. CELLiST™ F7 feed medium was specifically designed to work synergistically with CELLiST™ growth medium, including BASAL3, BASAL10, and CHO MX. However, it is not limited to these and can be used together with any commercially-available medium.



## Key Features of CELLiST™ F7:

- Employing Ajinomoto Group's proprietary cysteine-stabilization technology, high levels of readily available cysteine are achieved, allowing increased culture performance, while maintaining a hassle-free, single-agent feeding process at neutral pH.
- Amino acids and other medium components are optimized for increased process performance using cutting-edge 'Digital Twin' technology, through collaboration with the Bioprocess Technology Institute (BTI, A\*STAR).

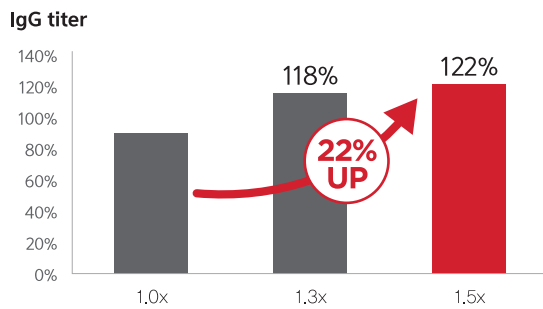
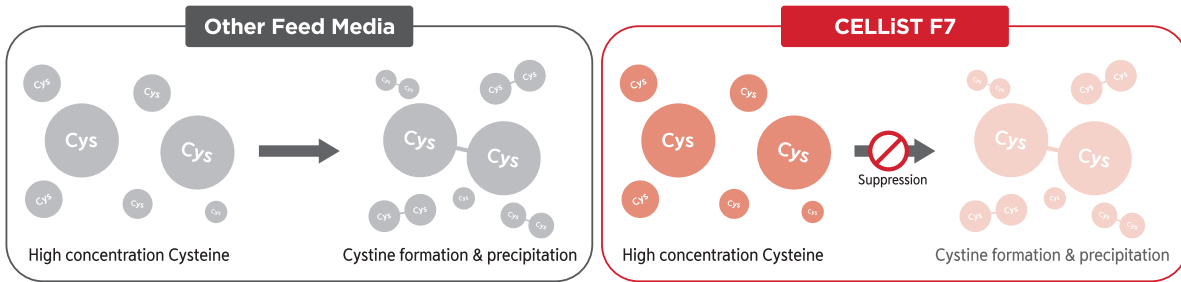
## Properties:

- Completely chemically-defined, protein-free, animal origin-free medium.
- Single-agent feed, easily dissolved at neutral pH, without the hassle of separated feed sources.
- Suitable for all CHO cell lines, including CHO-K1, CHO-GS, CHO-DG44 and CHO-S.
- CELLiST™ F7 can be combined with any commercially-available growth medium (although for best results, it is recommended to use CELLiST™ F7 in combination with CELLiST™ basal media).



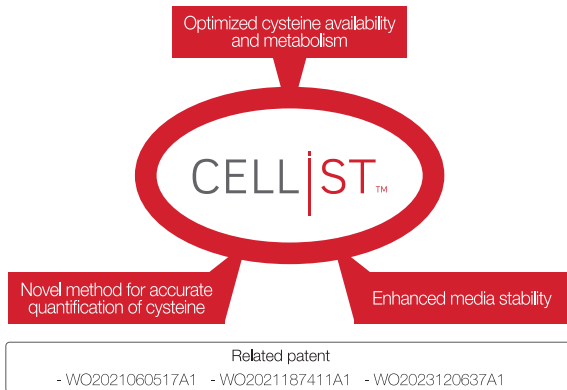
# Background:

- CELLIST™ F7 feed media employs Ajinomoto Group's proprietary cysteine-stabilization technology, ensuring high concentrations of cysteine are continuously available for your CHO cells throughout the culture, leading to greatly improved performance. This is achieved by drastically reducing the naturally-occurring oxidation of L-Cysteine to L-Cystine, which is easy to precipitate. This technology leads to increase in the amounts of available cysteine, while also improving stability and shelf-life.

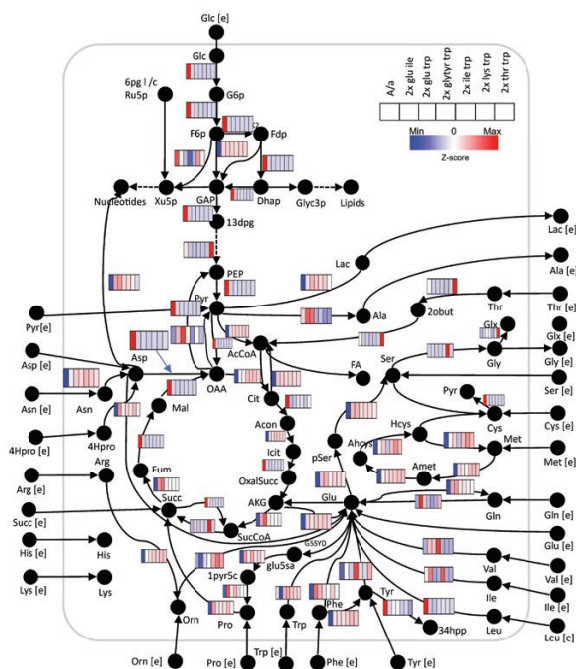


Above figure shows >20% increase in performance following 50% increase in available cysteine (1.5x compared to 1x) (ambr15® culture data, CHO-K1 cell line).

- Optimization of amino acids formulation through joint development with A\*STAR's Bioprocess Technology Institute.



Cutting-edge 'Digital Twin' and AI technologies were employed for optimizing metabolic flux in key pathways, such as the TCA cycle, leading to significant improvement in specific productivity and culture growth, and reduced levels of toxic by-products.



## Specifications

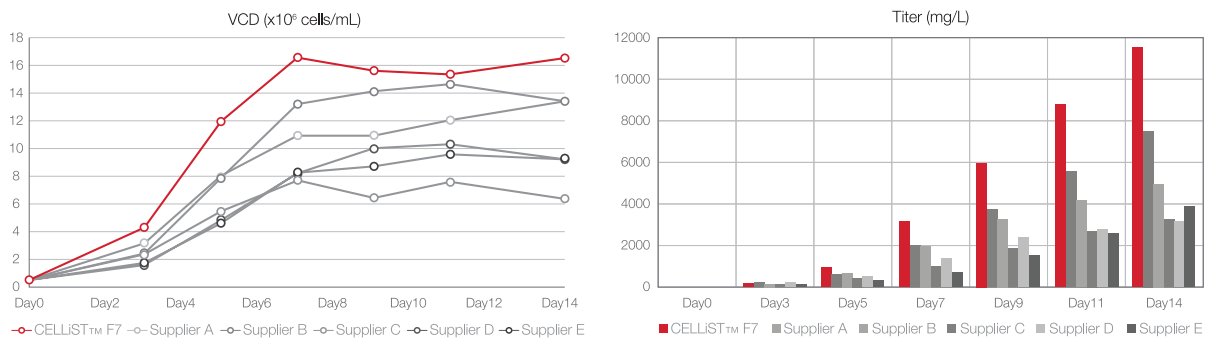
Model No.	F7
Format	Powder
Concentration	120.0 g / L
With Additives	None
Without Additives	L-Glutamine · D-Glucose · Insulin and other growth factors
Storage Condition	2°C to 8°C, dark and dry
Shelf Life	24 months
Item Description	Powder, chemically defined and animal derived component free medium

## Media Performance

CELLIST™ F7 feed media, together with CELLIST™ BASAL3 growth media, was compared against popular commercial media brands, as shown below. Ambr15® fed-batch process was employed. CELLIST™ F7 feed media was added from Day 3 to Day 11, every other day, at 6% (v/v). For other media brands, manufacturers' feeding recommendations were followed. Cell lines: CHO-K1 and CHO-GS.

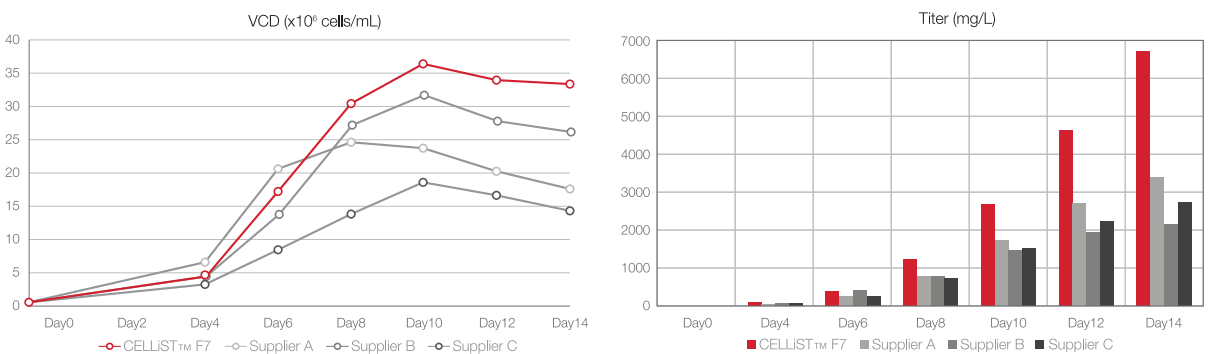
### CHO-K1 cell line

Fed-batch culture of CELLIST™ BASAL3 with CELLIST™ F7



### CHO-GS cell line

Fed-batch culture of CELLIST™ BASAL3 with CELLIST™ F7



# Liquid Medium Preparation

## Storage conditions:

Before liquid preparation, store powder media in a dark and refrigerated place (2–8°C), away from high humidity. After liquid preparation, store in a dark and refrigerated place (2–8°C) and use within 1 month.

## Instructions for preparation of liquid medium:

Table 1: Various parameters for preparation of 0.2 L of feed medium (with and without glucose addition):

Glucose concentration	Powder weight	Glucose weight	8N NaOH solution to be added	Total added water	pH*	Osmotic pressure* (5-fold dilution)	Total solution weight	Specific gravity (Room temperature)
0 g/L	24.0 g	0 g	2.0 mL (2.66 g)	181 mL (181 g)	6.6–7.0	205–225 mOsm/kg	208 g	1.04
70 g/L	24.0 g	14 g	2.2 mL (2.93 g)	173 mL (173 g)	6.6–7.0	280–300 mOsm/kg	214 g	1.07

\*Reference value

1. Prepare a suitable container and stir bar (magnetic bar). To ensure sufficient stirring, we recommend a container with a capacity of about 2–3 times the total prepared volume. When preparing on a weight basis, measure the weight of the container and the stir bar.
2. Fill the container with about 70% volume (140 mL) of cell culture-grade water (room temperature).
3. The total amount of this pouch (24.0 g) should be added to the container. Place a small amount of cell culture-grade water in the pouch to wash the remaining product into the container.
4. Add glucose as needed.
5. Stir for about 30 minutes.
6. Referring to Table 1, add 8N NaOH solution at the required amount. When adding glucose at amounts other than those in the table, please adjust the pH to 6.6–7.0 using 8N NaOH solution.
7. Stir for at least 30 minutes or until the powder is completely dissolved.
8. Make sure the pH is between 6.6–7.0. If the pH is lower than 6.6, add 8N NaOH solution to adjust to 6.6–7.0. If the pH is higher than 7.0, the stirring time should be extended to allow complete dissolution of the medium components.
9. Adjust to the final volume (200 mL) with cell culture-grade water and stir until the solution is clear for approximately 15 minutes. Volume adjustments can also be done by weighing (see table above).
10. Check pH and osmotic pressure. Osmotic pressure is measured by x5 dilution.
11. Under aseptic conditions (e.g., in biosafety cabinet), filter sterilization using a filter with a pore size of 0.20–0.22 µm.
12. Store in a refrigerated (2–8°C), dark environment until use.

## Usage:

- This product is a cell culture medium used for research applications. Do not use it for any other purpose.
- For use in manufacturing, and for any other inquiries, please contact us (details at the bottom of this document).

## Recommended fed-batch culture conditions in shake flasks:

- (1) Prepare liquid media following CELLIST™ media preparation instructions (see above).
- (2) Inoculate cells to 30 mL of basal media at  $0.5 \times 10^6$  cells/mL in 125 mL vented cap shake flask.
- (3) After inoculation, perform measurements to confirm appropriate cell density and viability.

## Recommended feeding strategy:

- Feeding volumes in the table below are stated as % (v/v) from the total initial volume.
- It is recommended to start feeding from Day 3, with daily additions of feed. If it is difficult to perform daily feed additions, then every other day is possible (see table below).
- Feeding volume depends on cell line characteristics. For high-productivity cell lines (titer >5 g/L), larger feeding volume may be required. See table below for information.
- Make sure to measure glucose concentration daily and top up glucose separately to maintain a concentration of 2–6 g/L (glucose consumption rate varies depending on cell line).
- For getting the most out of your cell line, it is recommended to optimize feeding volume by testing multiple feeding volumes (e.g., 2%, 4%, 6% every other day).

Example for recommended feed addition:

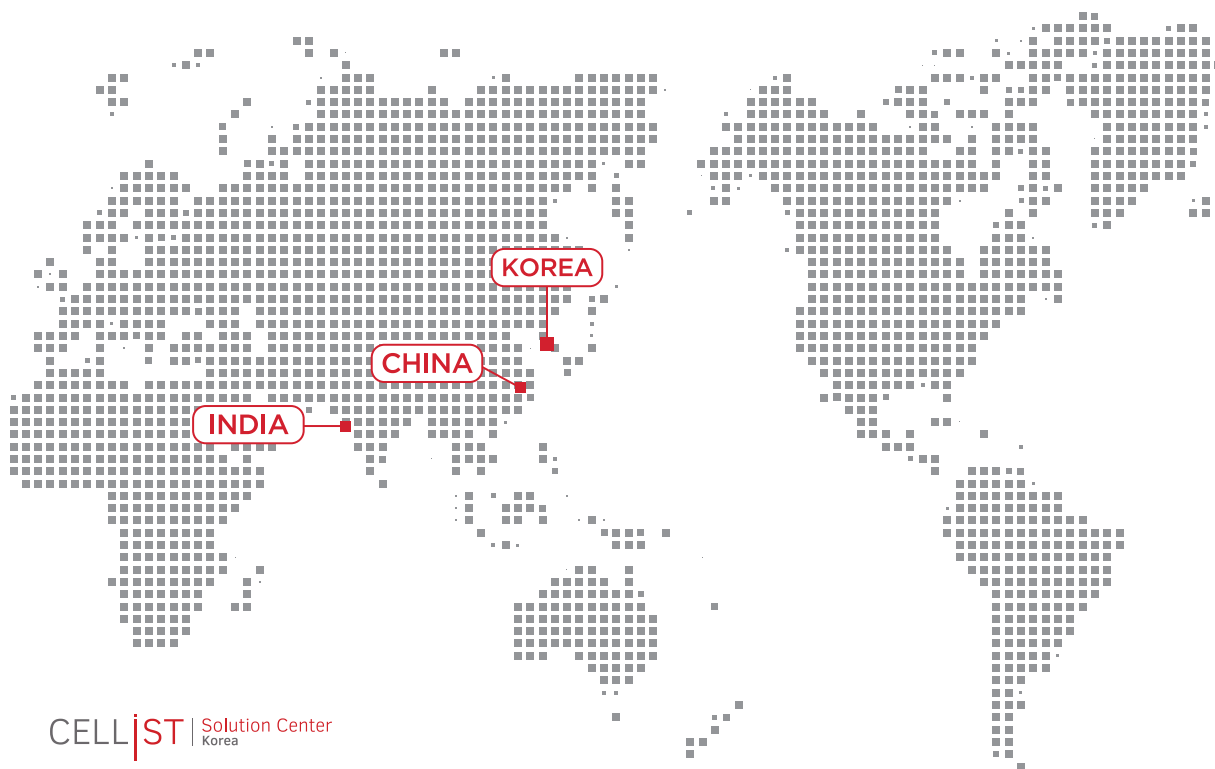
Feed	Cell type	Cultivation day																		
		1	2	3	4	5	6	7	8	9	10	11	12	13	14					
F7 (%v/v)	High titer (≥5 g/L)			3%	3%	3%	3%	3%	3%	3%	3%	3%	3%	3%						
F7 (%v/v)	Low titer (<5 g/L)			2%	2%	2%	2%	2%	2%	2%	2%	2%	2%	2%						
Glucose		Measure daily and maintain at 2–6 g/L																		

Feed	Cell type	Cultivation day																		
		1	2	3	4	5	6	7	8	9	10	11	12	13	14					
F7 (%v/v)	High titer (≥5 g/L)			6%	6%	6%	6%	6%	6%	6%	6%	6%	6%	6%						
F7 (%v/v)	Low titer (<5 g/L)			4%	4%	4%	4%	4%	4%	4%	4%	4%	4%	4%						
Glucose		Measure daily and maintain at 2–6 g/L																		



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