

Cellvento® ModiFeed Gal- COMP

Chemically defined Cell Culture Feed designed to reduce Galactosylation

Product Description

Cellvento® ModiFeed Gal- is a chemically defined feed formulation containing only components of non-animal origin. The product is intended for use in the development and manufacturing of monoclonal antibodies (mAbs) and next-generation biopharmaceuticals in Chinese hamster ovary (CHO) cell-based expression systems. For ease of use, the product is supplied as a compacted dry powder – a convenient to handle and hydration-friendly format.

Cellvento® ModiFeed Gal- is a **complete feed**. It is **highly concentrated**, one-part, pH neutral and designed to be added to replenish depleted nutrients required for cellular function and to extend the production phase in fed-batch, while simultaneously **reducing galactosylation** on the protein of interest.

Application

Glycosylation is known to have effects on the biological activity, solubility, serum half-life, and safety of therapeutic proteins. In a fed-batch process, cells are initially grown in a lean production medium followed by routine feeding to achieve the desired product yield. Control of protein glycosylation processes, such as galactosylation, while maximizing productivity is key for successful mAb development and production.

While novel product developments aim to achieve high galactosylation, in biosimilar development a given profile has to be obtained to mimic the original protein. As a result, having full control of the galactosylation levels and the specific amount of galactose on a glycan is critical. Cellvento® ModiFeed Gal- Feed was developed to increase G0(F) and decrease G1(F)/G2(F) while maintaining or improving product titer. Besides minimizing galactosylation, the galactosylation profile can be tailored as needed when combining Cellvento® ModiFeed Gal- Feed with Cellvento® ModiFeed Prime Feed and administering the combination as a single feed (see section B. *Initial galactose optimizing evaluation* for more details).

This product is intended for research or manufacturing but not for human or therapeutic use.



Target:
maximized
terminal GlcNAc

Benefits of the Cellvento® ModiFeed Gal- Feed include:

- Complete feed minimizing galactosylation.
- Chemically defined, non-animal origin components – providing increased process control and decreased complexity.
- Compacted feed – reducing the complexity of hydration, storage, and feeding.
- Hydrated feed is stable at room temperature – no temperature cycling required during processing.
- Highly concentrated – allowing for a reduced volume of feed, thereby increasing product yield.
- No hydrolysates, phenol red, or 2-mercaptoethanol – ensuring batch-to-batch consistency.
- No glucose – enabling greater control of glucose concentration during a fed-batch process.

Storage

Store product in original packaging at 2-8 °C protected from light.

Do not use after expiration date. Avoid opening and closing container multiple times.

Shelf life is provided with the CoA or shelf-life information sheet.

Reconstitution method – Cellvento® ModiFeed Gal- COMP Feed

1. Slowly add 143.63 g/L of compacted powder to 85% of final volume Milli-Q® or similar cell culture grade water (18 – 25 °C) in an appropriately sized container.
2. Rinse weighing vessel as necessary to remove remaining compacted powder.
3. Vigorously mix for at least 10 minutes, solution will still be slightly turbid.
4. Using a calibrated vessel adjust to 100% volume (QS) with Milli-Q® or similar cell culture grade water (18 – 25 °C).
5. Vigorously mix for at least 90 minutes until solution is clear.
6. Measure final pH: Expected pH 6.7 ± 0.3 .
7. Measure final osmolality: Expected osmolality $1,300 \pm 75$ mOsm/kg.
8. Immediately filter using a sterilizing-grade filter ($\leq 0.22 \mu\text{m}$). Filter recommendations are provided below.
9. Store reconstituted Cellvento® ModiFeed Gal- Feed protected from light at 2-8 °C for up to 90 days or at room (18-25 °C) temperature for up to 30 days.

Note: This feed does NOT contain glucose.

Recommended feeding strategy

Cellvento® ModiFeed Gal- has been developed to use as a feed in combination with either the production media Cellvento® 4CHO or EX-CELL® Advanced CHO but the use is not limited to these combinations. As with most upstream bioprocesses, optimization of feed volumes and timing of feed administration should be empirically determined on a process and cell line specific basis to maximize performance.

Cellvento® ModiFeed Gal- Feed, used in conjunction with Cellvento® 4CHO or EX-CELL® Advanced CHO media, is recommended to be fed at between 17.5% - 22.5% total feed, depending on the demands of the clone(s) tested.

A. Initial feeding evaluation

The use of Cellvento® ModiFeed Gal- **as sole feed to minimize galactose on glycans** of mAbs is described in the instruction below (see section A. *Suggested initial feeding evaluation*).

B. Adjustment option

To target a specific glycosylation profile and obtain a desired galactose content on the protein of interest, Cellvento® ModiFeed Gal- can be used **as part of a mixed feed to target a specific glycosylation profile** in combination with Cellvento® ModiFeed Prime. (See section B. *Initial galactose optimizing evaluation* for more detail).

Parameter	Recommended range for evaluation
Cellvento® ModiFeed Gal- COMP	2.5% – 7.5% (v/v) per feed
Frequency	48-72 hour feed interval
Glucose	4 – 8 g/L monitor, maintain and adjust accordingly

A. Suggested initial feeding evaluation

To identify the optimum approach for the clone(s) being tested, initial evaluation should consist of 17.5% and 22.5% total feed. A low percentage has the potential to underfeed while a high percentage has the potential to overfeed. Unless the general demands of the clone(s) are already known, during initial evaluation it is suggested to test both a low and high total feed percentage.

Important considerations

- Initiate the feeding only when viable cell density is $\geq 2 \times 10^6$ cells/mL and no earlier than day three (to avoid over-feeding).
- Maintain supplementation with feed supplements and glucose until culture viability is less than 80%.
- Terminate and harvest cultures when viability drops below 70%.

Culture Day	Total Feed	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	Total Feed (%)
Cellvento® ModiFeed Gal- COMP (% v/v)	Low				3		3		5.5			3		3			17.5
Cellvento® ModiFeed Gal- COMP (% v/v)	High				4		4		6.5			4		4			22.5

If preferred, the schedule can be adjusted to support a more regular feed interval:

Culture Day	Total Feed	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	Total Feed (%)
Cellvento® ModiFeed Gal- COMP (% v/v)	Low				3.5		3.5		3.5		3.5		3.5				17.5
Cellvento® ModiFeed Gal- COMP (% v/v)	High				4.5		4.5		4.5		4.5		4.5				22.5

Once a preference for a lower or higher total feed percentage is established, optimization through further increasing or decreasing the total feed can be evaluated.

The suggested initial feeding evaluation is designed to support a low seed ($2-5 \times 10^5$ cells/mL) 14 day fed-batch process. If viability is still high late in culture, longer fed-batches can be supported. Additional feeding may be required. Continue with similar feeding frequency and percentages until viability drops below 80%.

A high seed fed-batch ($2-10 \times 10^6$ cells/mL) may require adjustment in feeding schedule to support higher biomass earlier in culture and provide more total feed. Feeding can be initiated as early as day 2 to support higher biomass early in culture. High seed may offer an option for shortening the duration of longer fed-batches or for achieving increased production over the same 14 days.

B. Evaluation and Optimization of the galactose level.

Cellvento® ModiFeed Gal- Feed should be used at 100% to minimize galactosylation on terminal glycans.

If targeting a specific galactosylation profile is required, Cellvento® ModiFeed Gal- and Cellvento® ModiFeed Prime can be mixed. This method will decrease the galactose to a desired target level because the concentration of galactose-decreasing modulators is reduced and thus users can target a specific galactosylation profile. Testing will be required to optimize the amount of Cellvento® ModiFeed Gal- COMP Feed to Cellvento® ModiFeed Prime Feed. The same feed regiment should be employed as described in section A. *Suggested initial feeding evaluation.*

Prepare both feeds per each product's hydration instructions. After sterile filtration, the two complete

feeds can be mixed in a sterile container and should be protected from light. The feed created is complete and can be administered throughout a fed-batch as a single feed.

Initial testing mix suggestion for Cellvento® ModiFeed Prime and Cellvento® ModiFeed Gal- Feed is shown in the example graph below:

Amount (%) Cellvento® ModiFeed Prime Feed	Amount (%) Cellvento® ModiFeed Gal- Feed
0	100
25	75
50	50
75	25
100	0

When Cellvento® ModiFeed Prime Feed is mixed with Cellvento® ModiFeed Gal- Feed, a titration of the impact on galactose is observed (see **Figure**). The shift is dependent on the baseline achieved with the basal medium.

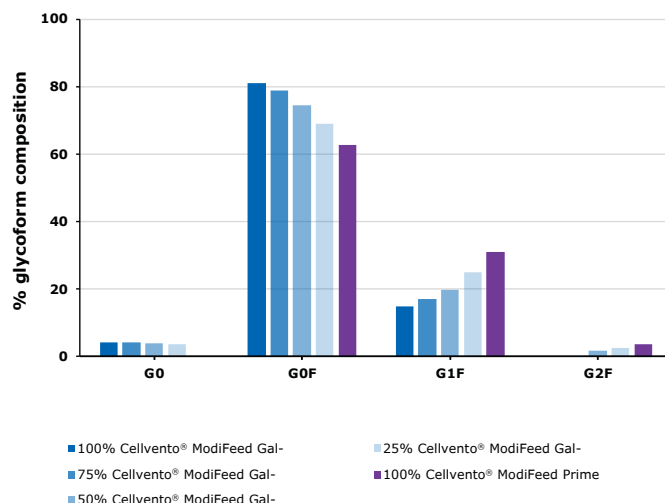


Figure: Glycosylation profile of CHOZN® Clone grown in EX-CELL® Advanced CHO Medium on day 14 with a varying composition of fed-batch culture feed.

Ordering Information

Cat. No.	Description	Application	Pkg. Size	Equivalent (L)
1.04648.0001	Cellvento® ModiFeed Gal- COMP	Low galactosylation feed	143.6 g	1
1.04648.0005	Cellvento® ModiFeed Gal- COMP	Low galactosylation feed	0.718 kg	5
1.04648.0050	Cellvento® ModiFeed Gal- COMP	Low galactosylation feed	7.180 kg	50

Related Products

Cat. No.	Description	Application	Pkg. Size	Equivalent (L)
1.03795	Cellvento® 4CHO COMP	Fed-batch medium	Multiple	1, 10, 100
24366C	EX-CELL® Advanced CHO	Fed-batch medium	Multiple	1, 10, 100
14366C	EX-CELL® Advanced CHO	Fed-batch medium	1000 mL	1
1.04132	Cellvento® ModiFeed Prime COMP	Fed-batch feed	Multiple	1, 5, 50

Filters

The following sterilizing-grade filters are recommended for use with Cellvento® ModiFeed Gal- COMP Feed:

Organism Removal	Bacteria Removal	Mycoplasma & Bacteria Removal
Volume (L)	Millipore Express® SHC	Millipore Express® SHR with Prefilter
5-50	KHGES015FF3	KHVES015FF3
500	KHGES03TT3	KHVES05TT1

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