

Technical Note

Guidelines for Fed-Batch Evaluation & Feeding Manner Optimization

Cell culture growth and protein production are highly dependent on cell line characteristics. Therefore, for achieving best results, feeding manner should be optimized specifically for each cell line. Here, we provide general guidelines for fed-batch evaluation and for optimization of feeding manner.

Cell culture conditions:

- 1) Seeding density: 3-5 x10⁵ cells/mL.
- 2) Supplements: Should be added depending on cell line (for e.g., 2-6 mM L-Glutamine, or as required).
- 3) Incubator: 37°C; 5% CO₂, humidified.
- 4) Glucose: should be added either separately or together with feed, as desired. Maintain > 4g/L, or according to cell line glucose needs.

Feeding manner optimization:

In general, feeding should be performed at volume range of 2%-6% (v/v of total liquid volume), every two days, starting at day 3 or 4. In order to find the optimal feeding regime, we recommend evaluating feeding pattern according to the table below.

Culture Day										Total Vol.
3	4	5	6	7	8	9	10	11	12	
2%		2%		2%		2%		2%		10%
4%		4%		4%		4%		4%		20%
6%		6%		6%		6%		6%		30%
	2%		2%		2%		2%		2%	10%
	4%		4%		4%		4%		4%	20%
	6%		6%		6%		6%		6%	30%

Example Data:

Below is culture data including viability, viable cell density (VCD) and IgG titer for a CHO-K1 cell line. Culture was performed using ambr15[®] microbio reactor system, and feed was added at day 4, 6, 8, 10 and 12. It can be seen that feeding volume has strong effect on culture growth and productivity, and, in this particular cell line, 2% feeding was shown to result in optimal culture performance.

