

HyClone™ CD BEVS complete medium

CELL CULTURE MEDIA AND SUPPLEMENTS

HyClone™ CD BEVS complete medium is a chemically defined (CD), animal-derived component-free (ADCF), serum-free insect cell culture medium developed to support robust growth and high-yield production in baculovirus expression vector systems (BEVS). Designed for ease of adoption and consistent performance, the medium drives high-density Sf9 insect cell growth to deliver high viable cell densities (VCD) and product titers across key applications, including virus like particle (VLP) production, recombinant adeno-associated virus (rAAV) manufacturing, recombinant protein expression, and other BEVS workflows. This medium supports scale-up from shake flasks to bioreactors and does not require supplementation or complex adaptation strategies.

Key features of HyClone CD BEVS complete medium:

- Chemically defined, hydrolysate-free, serum-free and ADCF formulation.
- Supports direct adaptation from other insect cell media.
- Delivers higher VCD and titers than legacy HyClone insect cell media (SFM4Insect and SFX-Insect cell culture media), as well as Gibco ExpiSf CD medium and Merck EX-CELL CD insect cell medium.
- Maintains high cell viability (> 90%) throughout extended culture durations.
- Performs consistently across packaging formats and culture scales.
- Optimized for BEVS-based recombinant protein and viral vector production.



Fig 1. HyClone CD BEVS complete medium is available in liquid or powder form in pack sizes suitable for small-volume cell culture as well as large-scale bioprocessing applications.

Adaptation data

CD BEVS enables straightforward, direct adaptation from cells cultured in other commercially available media without the need for stepwise adaption or supplementation while preserving viability and growth performance. We evaluated HyClone CD BEVS complete medium when used for direct adaptation of Gibco Sf9 insect cells from legacy HyClone SFX-Insect and SFM4Insect media as well as from commercially available ExpiSf CD and EX-CELL CD insect media. As shown in Figure 2, Sf9 cultures directly adapted into CD BEVS complete medium maintained high viability and robust growth kinetics following adaptation, with peak VCD values comparable to or exceeding those observed in the incumbent media.

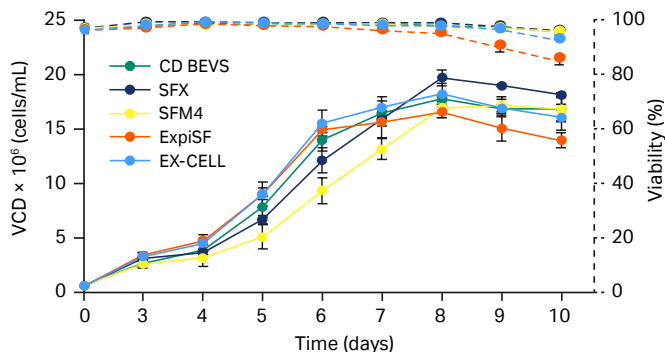


Fig 2. Adaptation of Sf9 cells from various insect cell culture media into CD BEVS complete medium. Sf9 cells cultured in HyClone SFX Insect, SFM4Insect, Gibco ExpiSf CD, or Sigma EX-CELL medium were directly adapted into HyClone CD BEVS complete medium. Following three passages, cultures were grown in batch mode and viable cell density (VCD) and viability were monitored over time. Cells continuously maintained in CD BEVS complete medium served as the control. Data represent mean $\pm$ SD ($n = 3$ biological replicates).

Growth performance data

We also compared growth and productivity of Sf9 cells cultured in legacy hydrolysate-based HyClone media, third-party CD insect cell media, or HyClone CD BEVS complete medium. As shown in Figure 3, Sf9 cells cultured in this medium achieved higher peak VCD values than those grown in HyClone SFX-Insect, HyClone SFM4Insect, ExpiSf CD, or EX-CELL CD media, while maintaining high culture viability throughout the growth phase.

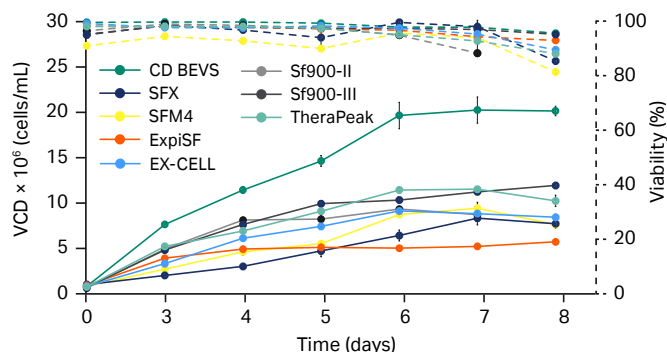


Fig 3. Batch growth promotion study ($n=3$ for each condition) seeded at 0.75×10^6 vc/mL showed faster early phase growth (22.2 ± 2.3 hr PDT), higher peak VCD ($2.0 \times 10^7 \pm 0.66$ vc/mL), and superior late-phase viability in CD BEVS complete medium. Dunnett's test demonstrated significantly improved growth relative to all other conditions ($p < 0.0001$; viability $p < 0.01$).

Historically, insect cell culture media have relied on complex components such as yeastolates or protein hydrolysates to support performance. While effective, these undefined components introduce batch-to-batch inconsistencies and generate challenges in regulatory submissions.

The medium supports consistent growth kinetics, which allows for ease-of-use, consistent growth, and reliability when scaling up to a production run (Fig 4).

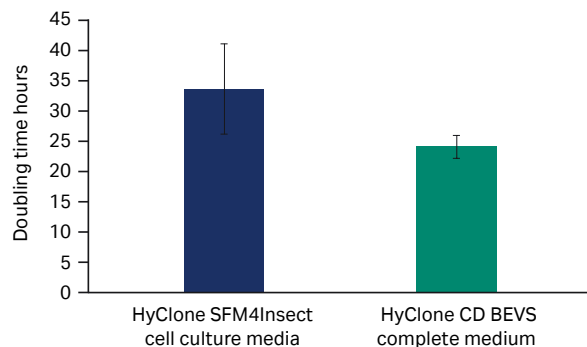


Fig 4. Growth kinetics of multiple batches of SFM4Insect ($n = 22$; population doubling time = 33.6 ± 7.5 h) and CD BEVS complete medium ($n = 6$; population doubling time = 24.0 ± 1.9 h). Cells cultured in CD BEVS complete medium exhibited a faster population doubling time and lower batch-to-batch variability compared with SFM4Insect.

BEVS performance data

We also assessed the medium in post-infection studies of Sf9 cells after infection with IgG expressing baculovirus under equivalent conditions comparing cells grown in different media. Cells showed higher IgG titers when cultured and infected in HyClone CD BEVS complete medium than in HyClone SFM4Insect media, HyClone SFX-Insect media, ExpiSf CD media, or EX-CELL CD media (Fig 5), indicating improved productivity under equivalent culture and infection conditions.

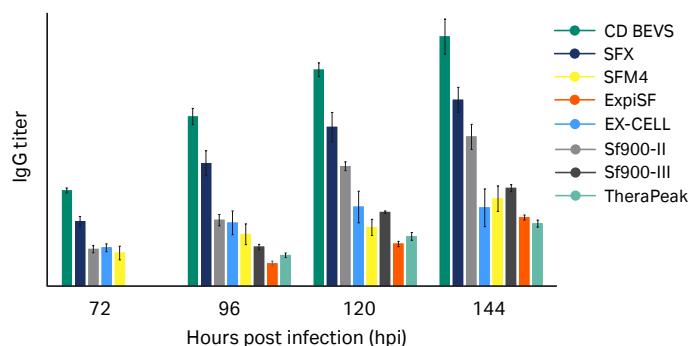


Fig 5. IC-BEVS IgG expression yielded faster kinetics and higher titers in CD BEVS complete medium versus all other conditions ($p < 0.0001$). Sf9 cells at 3×10^6 cells/mL were infected with a multiplicity of infection (moi) of 0.1. IgG production was monitored for 6 d post-infection. post-infection IgG titers are in cells cultured in HyClone CD BEVS complete medium, HyClone SFX-Insect (SFX), HyClone SFM4Insect (SFM4), ExpiSf CD, Thermo Sf900-ii, Thermo Sf900-iii, Lonza TheraPeak SfAAV, or EX-CELL CD media. ($n = 3$ biological triplicates, error bars show ± 1 SD).

Scale-up data

We assessed HyClone CD BEVS complete medium in scale-up from shake flask cultures to bioreactor systems. As shown in Figure 6, Sf9 cultures grown in a 2 L benchtop bioreactor achieved higher peak VCD values than shake flask cultures, while maintaining high viability across the cultivation period. We attribute this difference to the active process control that occurs in the bioreactor system, which may enhance performance in comparison to the minimally controlled flask environment. Nevertheless, the overall performance trends between the two cultivation formats were consistent. These data indicate that HyClone CD BEVS complete medium is suited for scalable BEVS workflows and supports robust culture performance under process control conditions typical of bioreactor operation.

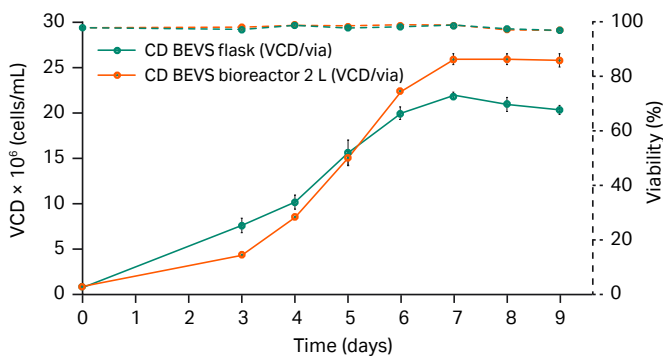


Fig 6. Sf9 culture performance in 50 mL Erlenmeyer flasks ($n = 3$ biological triplicates, error bars show 1 SD) vs a 2 L benchtop bioreactor ($n = 1$ single biological sample; error bars indicate technical replicates ± 1 SD). Cells were cultured in HyClone CD BEVS complete medium.

Specifications

- Chemically defined, ADCF, serum-free formulation.
- Optimized for Sf9 insect cell culture and suitable for Sf21, High Five cells, and BEVS applications.
- No supplementation required to achieve stated performance.
- Manufactured under ISO 13485-certified quality systems.

Product handling

Store medium at 2–8°C and protect from light. Powdered medium should be stored in a tightly sealed container to prevent moisture uptake. Liquid media are stable for up to 6 months, and powder media are stable for up to 12 months when stored as recommended. Note: product is undergoing real-time stability testing, and shelf life may be extended.

Custom production

Formulations and delivery systems can be customized to your specific process requirements or optimized to maximize process yields.

Rapid response production (RRP)

Our RRP program manufactures up to 200 L of liquid medium, or 20 kg of DPM of your custom prototype formulation within seven working days of your request. Use our RRP service to expedite the development and testing of custom buffers and process liquids for your biopharmaceutical manufacturing process.

Ordering information

Product	Size	Packaging	Product code
HyClone CD BEVS complete medium, liquid medium	1000 mL	Bottle	SH31204.01
	1 L	Bag	SH31204.02
	5 L	Bag	SH31204.03
	10 L	Bag	SH31204.04
	20 L	Bag	SH31204.05
	50 L	Bag	SH31204.06
	100 L	Bag	SH31204.07
	200 L	Bag	SH31204.08
HyClone CD BEVS complete medium, dry powder medium	5 L	HDPE bottle	SH31205.01
	10 L	HDPE bottle	SH31205.02
	20 L	HDPE bottle	SH31205.03
	50 L	HDPE bottle	SH31205.04
	100 L	Poly bag/pail	SH31205.05
	500 L	Poly bag/pail	SH31205.06
	1000 L	Poly bag/pail	SH31205.07



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