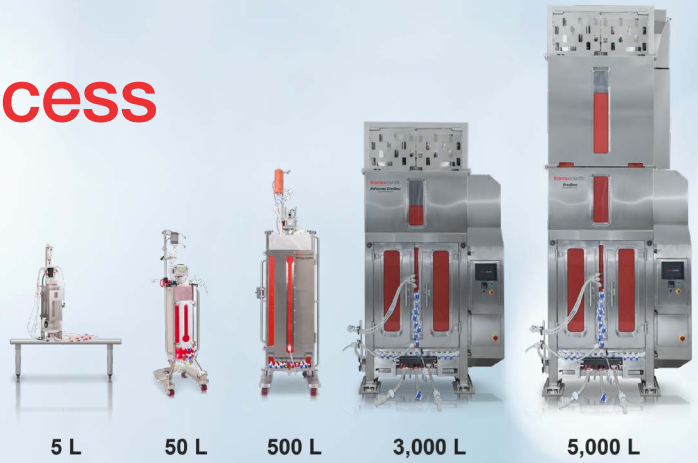
Single-use bioprocessing

# Demonstrating bioprocess scalability through platform consistency

## A 1,000-fold scale-up in the DynaDrive S.U.B.




### Introduction

The Thermo Scientific™ DynaDrive™ Single-Use Bioreactor (S.U.B.) product line enables a unified path from bench to production through a consistent design philosophy that is carried from 1 L working volume through 5,000 L. Variables and unknowns during tech transfer are reduced across scales by using the same Thermo Scientific™ Aegis™ 5-14 film, laser drilled-hole spargers, multi-impeller drivetrain, vessel aspect ratio, and fluid-contact materials. By this platform design, predictable mixing, mass transfer, and cell culture performance are supported, and control strategies are more readily translated.

The 5 L DynaDrive S.U.B. is positioned to be used by process development and production groups as an advanced scale-up tool and scale-down model, useful for screening and developing processes and acceptance criteria, optimizing control strategies, making comparability assessments, and de-risking technology transfer to larger DynaDrive vessels.

Through platform consistency and demonstrated scalability, the DynaDrive product line can enable faster iteration at 1 L to 5 L working volumes, greater confidence in scale decisions, and more straightforward transfer of bench-established strategies to production-scale execution. Previously, comparable growth, viability, and product titers have been observed when scaling from 5 L to 50 L under straightforward, similar control conditions [1]. Now the approach is applied to scaling from 5 L to 5,000 L.

This case study demonstrates a straightforward, execution-friendly path from bench to production using a single primary anchor: constant power input per volume (P/V of ~20 W/m$^3$), with aeration and controls tuned to standard DO and pH targets. The objective isn't to claim universality for one scaling criterion, but to show how maintaining a simple, defensible anchor can translate performance across three scales on the same platform. As discussed previously [2], there are various approaches that can be used for scaling, each with limitations and considerations; the method selected for this case study is shown to be a viable starting point for process scaling across various orders of magnitude in the DynaDrive product line.

Methods and control parameters are detailed in the next section; briefly, the run leverages the platform's common materials, impeller system, and sparger design to avoid confounding geometry changes. The run also uses a common control ecosystem, namely the Thermo Scientific™ HyPerforma™ G3 family of bioprocess controllers and Thermo Scientific™ TruBio™ automation software at each scale.

Notable outcomes are that viable cell density and viability track closely across 5 L, 50 L, and 5,000 L; IgG accumulation profiles also align. Together, these results indicate that the constant P/V strategy for scaling via the DynaDrive S.U.B. helps provide sufficient mixing and mass-transfer capability to carry the process across a 1,000-fold scale difference without extensive re-optimization.

## Methods

Thawing and expansion of an IgG-producing CHO-K1 clone were performed using Gibco™ Efficient-Pro™ AGT™ Medium in shake flasks under standard procedures. Expansion proceeded in flasks until sufficient material was generated to inoculate a 50 L DynaDrive S.U.B. at full working volume with a targeted seeding density of 0.25 x $10^6$ cells/mL. Cell expansion continued in the 50 L DynaDrive S.U.B. for 4 days, at which point the cell density was sufficient to inoculate a 5,000 L DynaDrive S.U.B at approximately 280 L working volume with a targeted seeding density of 0.3 x $10^6$ cells/mL. After 3 days of expansion at this low volume (nearly 20:1 turndown ratio), there were sufficient cells to inoculate various n-stage cultures taking place in the 5 L, 50 L, and 5,000 L S.U.B.s. This approach demonstrated the effectiveness of scaling across bioreactors (Figure 1).

No probe installations were required, as each of the Thermo Scientific™ DynaDrive™ BioProcess Containers include pre-integrated single-use pH probes and DO probe adaptors. Integrated pH and DO probes were calibrated as necessary prior to inoculation of each vessel. The required amount of seed material was drained from the 5,000 L DynaDrive S.U.B. to inoculate the 5 L and 50 L S.U.B.s, each of which were previously filled with sterile medium sufficient to reach the target initial volume with the addition of the inoculum. Additional draining required to remove any excess cell material and provide the appropriate initial cell density in the 5,000 L S.U.B. took place, followed by addition of sterile medium to reach the target initial volume. On day 3 of the n-stage process, Gibco™ Efficient-Pro™ AGT™ Feed 1 started at a constant-flow addition rate of 2.25% of the current vessel volume per day, with the rate being updated daily. A 36% (w/v) glucose solution was also supplemented continuously, with the rate adjusted daily as needed to maintain a glucose concentration of 3 g/L. Gibco™ FoamAway™ Irradiated AOF Antifoaming Agent was used to control excess foam through a combination of periodic dosing and activation by the integrated foam probe. Daily process samples were taken from the reactors to monitor cell density, cell viability, cell size, metabolites, and IgG production. These fed-batch cell cultures terminated on day 14.

The Thermo Scientific™ HyPerforma™ G3Lab™ Bioprocess Controller was used for the 5 L DynaDrive S.U.B., while Thermo Scientific™ HyPerforma™ G3Pro™ Bioprocess Controllers were used for the 50 L and 5,000 L DynaDrive S.U.B.s. The operation and control parameters applied at each scale are provided in Table 1, with similar targets selected to illustrate straightforward scalability and process transfer within the DynaDrive platform.

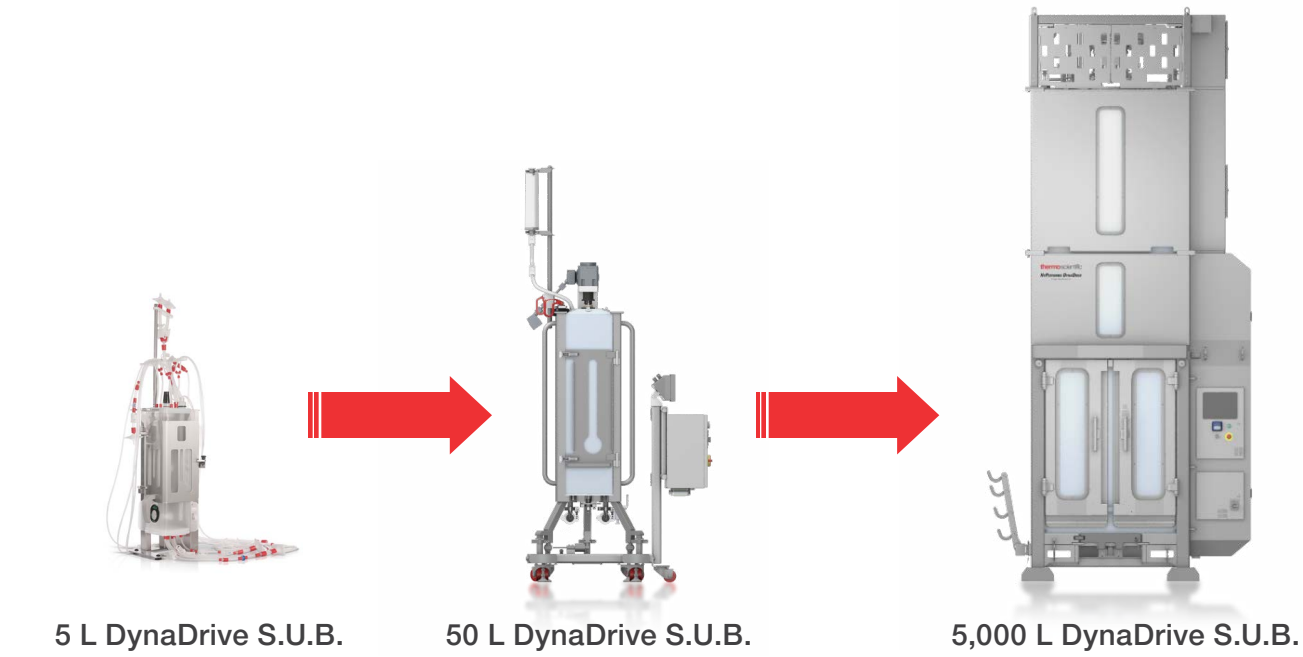



**Figure 1. Scale-up across a wide range of volumes with the DynaDrive S.U.B.**

**Table 1. Operation and control parameters.**

| Parameter | 5 L DynaDrive S.U.B. | 50 L DynaDrive S.U.B. | 5,000 L DynaDrive S.U.B. |
|---|---|---|---|
| Target initial/final volume | 3.75 L/5 L | 36 L/50 L | 3,600 L/5,000 L |
| Target seed density (x $10^6$ cell/mL) | 0.3 | 0.3 | 0.3 |
| Temperature set point (°C) | 37 | 37 | 37 |
| Agitation (rpm) | 230 | 105 | 46 |
| Power input per volume (W/$m^3$) | 20 | 20 | 20 |
| Tip speed (m/sec) | 0.63 | 0.59 | 1.18 |
| Drilled-hole sparger (DHS) configuration | 235 x 80 µm pores | 1,448 x 80 µm pores | 2,610 x 233 µm pores;<br>2 x 1,180 x 445 µm pores |
| Target glucose conc. (g/L) | 3 | 3 | 3 |
| Target feed addition rate | 2.25% of current vessel volume | 2.25% of current vessel volume | 2.25% of current vessel volume |
| Controller | HyPerforma G3Lab system | HyPerforma G3Pro system | HyPerforma G3Pro system |

DO and pH controls used in this case study, including set points, mass flow controller (MFC) scaling, and proportional-integral-derivative (PID) settings, were configured as provided in Tables 2 and 3. The output scaling for each of the gas species listed in the following tables indicates the limits imposed at time of run configuration, not to be interpreted as the maximum flow observed during the cultures.

**Table 2. Dissolved oxygen (DO) control parameters.**

| Parameter | | 5 L DynaDrive S.U.B. | 50 L DynaDrive S.U.B. | 5,000 L DynaDrive S.U.B. |
|---|---|---|---|---|
| DO set point | | 40 | 40 | 40 |
| DO PID | Gain | 0.1 | 0.1 | 0.5 |
| | Reset | 200 | 200 | 180 |
| $O_2$ | Controller output | 10–100% | 10–100% | 10–50%*<br>50–100%** |
| | Output scaling | 0–0.225 slpm | 0–2.25 slpm | 0–75 slpm*<br>0–100 slpm** |
| $N_2$ | Controller output | 0–10% | 0–10% | 0–10%** |
| | Output scaling | 0.025–0 slpm | 0.25–0 slpm | 25–0 slpm** |
| Air | Overlay | 0.25 slpm | 1 slpm | 20 slpm |
| | Controller output | 0–10% | 0–10% | 0–10%** |
| | Output scaling | 0–0.025 slpm | 0–0.25 slpm | 0–25 slpm** |

* Routed to the 233 µm pore size DHS.

** Routed to the 445 µm pore size DHS.

Table 3. pH control parameters.

| Parameter | | 5 L DynaDrive S.U.B. | 50 L DynaDrive S.U.B. | 5,000 L DynaDrive S.U.B. |
|---|---|---|---|---|
| pH set point | | 7.15 | 7.15 | 7.15 |
| pH PID | Gain | 1.6 | 1.6 | 1.6 |
| | Reset | 300 | 300 | 200 |
| pH deadband | High deadband | 0 | 0 | 0 |
| | Low deadband | 0.25 | 0.25 | 0.25 |
| | High gap | –0.15 | –0.15 | –0.15 |
| | Low gap | 0 | 0 | 0 |
| $CO_2$ | Controller output | –100 to 0% | –100 to 0% | –100 to 0%* |
| | Output scaling | 0.025–0 slpm | 0.25–0 slpm | 25–0 slpm* |
| Base | | Not enabled | Not enabled | Not enabled |

* Routed to the 233 µm pore size DHS.

## Results

From 5 L through 50 L and into the 5,000 L scale, the DynaDrive S.U.B.s yielded a consistent viable cell density (VCD) trajectory (Figure 2). Peak VCD for each culture lay between 53 x $10^6$ cells/mL and 55 x $10^6$ cells/mL and final VCD between approximately 33 x $10^6$ cells/mL and 36 x $10^6$ cells/mL. Cell viability also tracked similarly among each of the reactors and was consistent across all scales (Figure 3). Viability dropped from day 5 to day 14 in each culture, remaining above 90% through day 8 before dropping to a final viability of 65–75%.

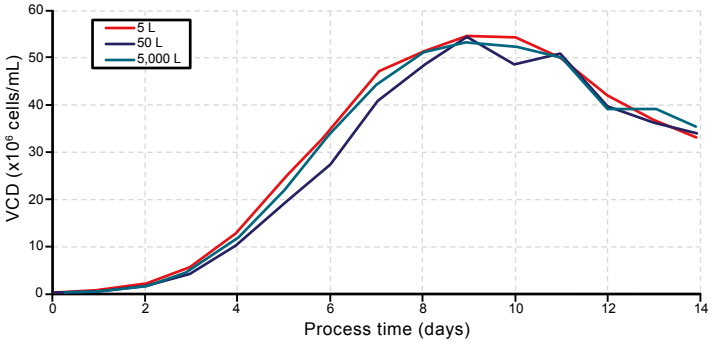



Figure 2. Viable cell density (VCD) for 5 L to 5,000 L DynaDrive S.U.B.s.

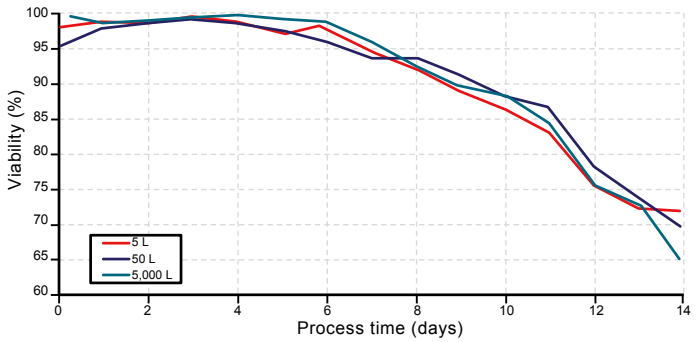



Figure 3. Viability for 5 L to 5,000 L DynaDrive S.U.B.s.

Overall pH and $pCO_2$ trends followed a similar pattern, though the 5,000 L DynaDrive S.U.B. did reach a slightly lower pH and slightly higher $pCO_2$ while the cultures were within the pH deadband region (Figure 4). Also, it may be observed that in this particular 5 L DynaDrive S.U.B. culture replicate, the online pH sensor experienced a slightly higher degree of sensor drift than in the other cultures, especially during the latter half of the culture. The instances of a sudden pH drop on days 0, 4, 7, 10, 11, and 12 are each due to a 1-point pH offset being implemented after an offline sample analysis. These pH offsets also resulted in some oscillation of the culture $pCO_2$ in the 5 L culture, resulting from $CO_2$ sparge modulation as a response to the pH control requirements. It is likely that were the pH measurement to be more stable, the culture $pCO_2$ would have remained closer to the midpoint of those oscillations, which aligns well with the 50 and 5,000 L DynaDrive S.U.B. culture $pCO_2$ measurements. Nonetheless, pH and $pCO_2$ remained within the expected and tolerable ranges for each of the cultures.

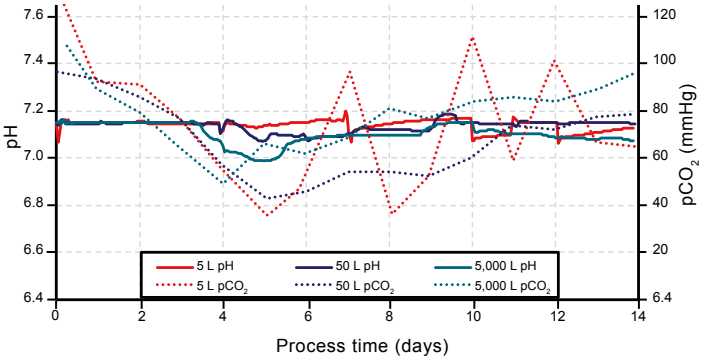



**Figure 4. pH and $pCO_2$ for 5 L to 5,000 L DynaDrive S.U.B.s.**

The DynaDrive S.U.B. offers efficient mixing and mass transfer. Even at a relatively modest power input per volume of 20 W/$m^3$, peak demand of sparged gasses (even during peak growth rate and peak cell density) remained low, with a combined gas flow rate typically at <0.05 VVM, far below the sparger design maximum of 0.15 VVM. The oxygen delivery to $CO_2$ stripping ratio was adequate in this study. If higher $CO_2$ stripping is needed, the DynaDrive S.U.B. is well within its broad operating range to allow for tuning of incoming sparged air/$O_2$ ratios. Other tuning options include control over bubble size ratios via sparger selection, or mass transfer adjustments via changes to agitation rate.

Lactate levels in the various cultures aligned reasonably, with the lactate consumption switch occurring over a similar period in each culture. Lactate peaked on day 4 in each culture after a primary accumulation phase, then decreased until about day 8 to day 9, then underwent a secondary accumulation phase until process termination (Figure 5). The 5,000 L DynaDrive S.U.B. culture peaked at 1.37 g/L lactate compared to 1.2 g/L in the 5 L S.U.B and 1.05 g/L in the 50 L S.U.B.

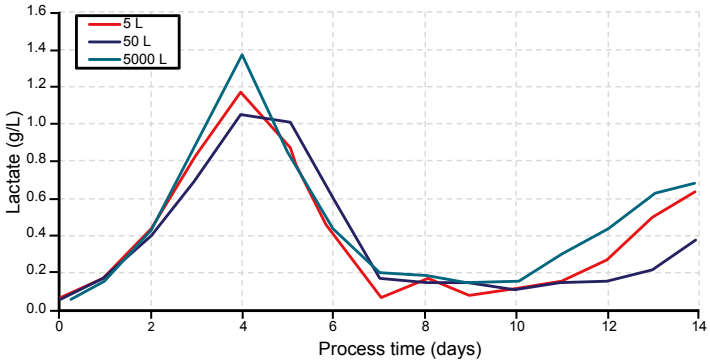



**Figure 5. Lactate levels for 5 L to 5,000 L DynaDrive S.U.B.s.**

IgG titer among the cultures increased at similar rates throughout, with the 5 L and 5,000 L DynaDrive S.U.B. cultures both accumulating ~3.5 g/L, compared to ~3.3 g/L in the 50 L culture (Figure 6). The apparent change in IgG accumulation rate occurring on day 9 is notably congruent across each reactor scale.

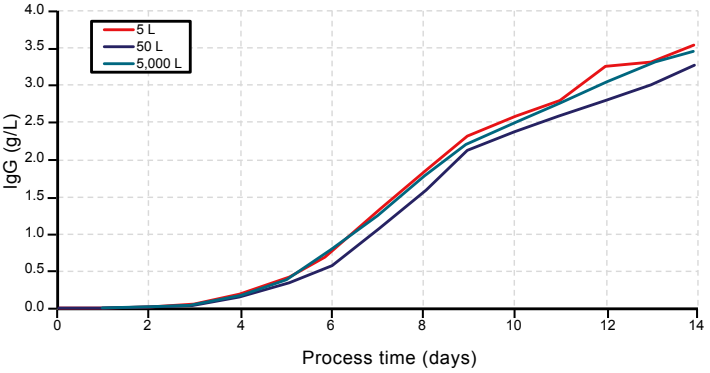



**Figure 6. IgG titer for 5 L to 5,000 L DynaDrive S.U.B.s.**

## Discussion and conclusion

The multi-impeller drivetrain of the DynaDrive S.U.B. product line offers robust power delivery throughout the working volume, helping to provide a reliable environment for scale-up. While no scaling criterion is perfect, operating this process at approximately 20 W/m$^3$ across scales was sufficient to simultaneously offer adequate mixing and efficient oxygen transfer. Furthermore, the reactor, port, and lineset design—along with the flexible working volume offered by the DynaDrive product—enabled an efficient and straightforward method to simultaneously inoculate three reactors across a 1,000-fold scale difference, all from the same seed train and without the use of or need for any additional reactors. The sparge design of the reactors also allowed for simple management of both oxygen demands and $CO_2$ stripping requirements as metabolic states of the cell cultures shift over time. Gassing requirements were modest, with plenty of design space yet available for more intensified or demanding processes.

In this case study, alignment of key performance indicators, including viable cell density, viability, and product titer, was maintained from 5 L through 50 L and up to 5,000 L under similar control strategies, illustrating that the development conditions established at 5 L translated predictably across scales of the DynaDrive platform. These results exhibit the utility of the 5 L DynaDrive S.U.B. for defining acceptance criteria, informing parameter targets at larger scales, and simplifying comparability assessments—thereby supporting increased confidence and efficiency during tech transfer to pilot and production operations.

## Ordering information

| Product | Cat. No. |
|---|---|
| Efficient-Pro AGT Medium | A5322303 |
| Efficient-Pro AGT Feed 1 | A5209102 |
| FoamAway Irradiated AOF Antifoaming Agent | A1036902 |
| HyPerforma G3Lab Controller | F100-U110-000 |
| HyPerforma G3Pro Controller | F100-P132-100, F100-P132-200 |
| DynaDrive Single-Use Bioreactor (5,000 L) | DDB5000.1011 |
| DynaDrive BioProcess Container (5,000 L) | SH31195.01 |
| DynaDrive Single-Use Bioreactor (50 L) | DDB0050.1011 |
| DynaDrive BioProcess Container (50 L) | SH31192.01 |
| DynaDrive Single-Use Bioreactor (5 L) | DDB0005.1010, DDB0005.1011, DDB0005.1020, DDB0005.1021 |
| DynaDrive BioProcess Container (5 L) | PROSUT00180-I, PROSUT00181-I |

## References


1. Thermo Fisher Scientific (2024) Part 2: Intuitive bioprocess scale-up from bench scale to pilot scale. Application note.
2. Thermo Fisher Scientific (2026) Criteria for effective bioprocess scale-up with the DynaDrive Single-Use Bioreactor. White paper.


Learn more at **thermofisher.com/dynadrive**