cytiva

Intensified upstream operations

With Xcellerex™ Automated Perfusion System (APS)

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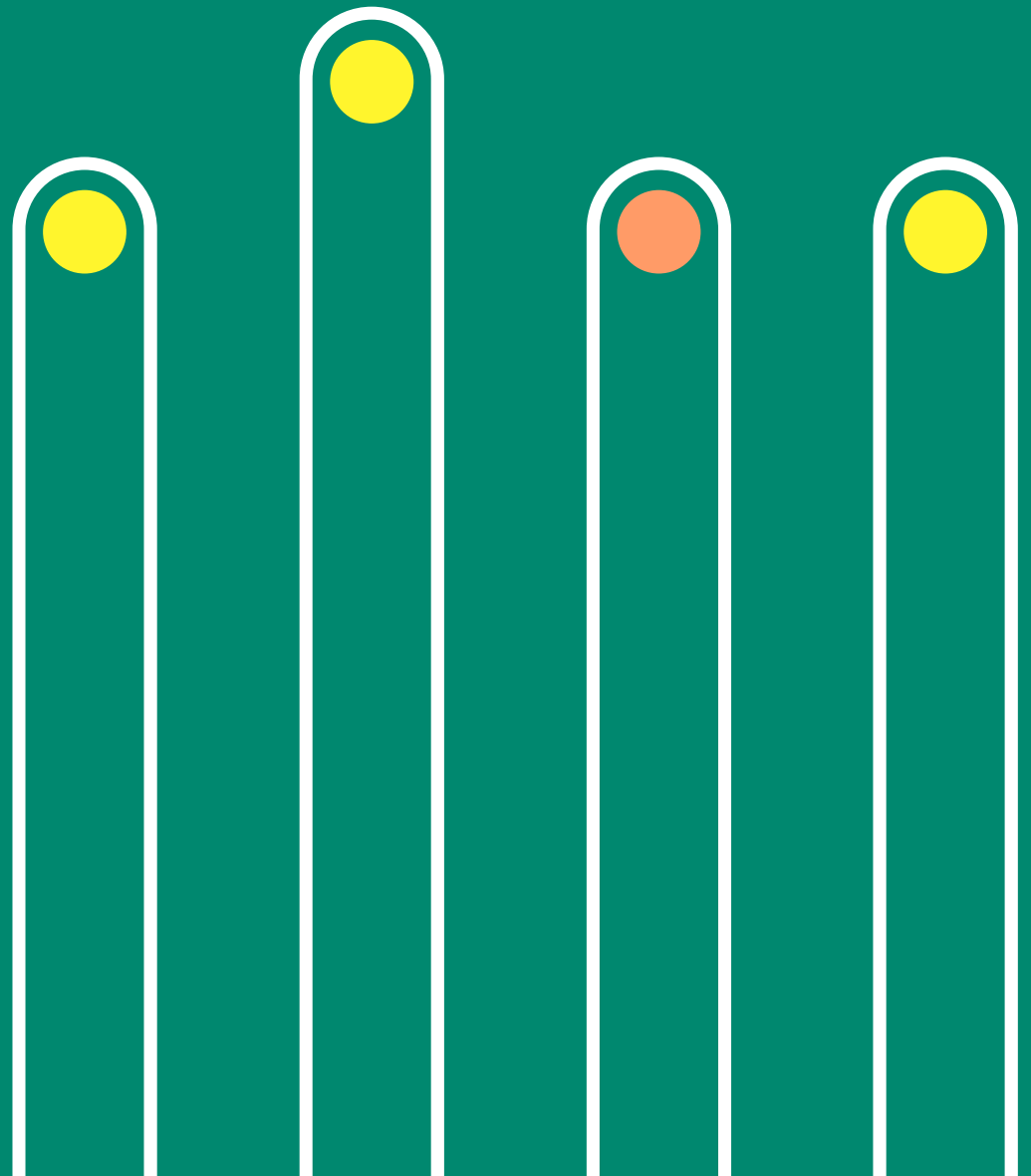

Agenda

1. Introduction
2. Case study: N-1 perfusion for fed-batch processes
3. Bring consistency to perfusion: introducing Xcellerex™ APS
4. Summary

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Introduction

Market trends and business drivers adapted from BPOG Technology roadmap

Market trends

Market growth
High demand

Number of drugs supplied

Global reach and emerging markets

New product classes
New treatment modalities

Personalized medicine

Uncertainty
Clinical efficacy, dose requirements

Product approvals, complex regulations

Demand, competition, and market share

Regional/political requirements

Cost pressure
Payer pressure on price

Biosimilars and competition

Cost of clinical failure

Escalating development costs

Business drivers

Facility flexibility
Facility design and scale

Multiproduct capability

Regional manufacture

Speed
Speed to clinic

Speed to build

Speed to market

Speed to supply

Quality
Product attributes and characterization

Comparability requirements

Quality/risk management

Cost of nonquality

Cost reduction
Development costs

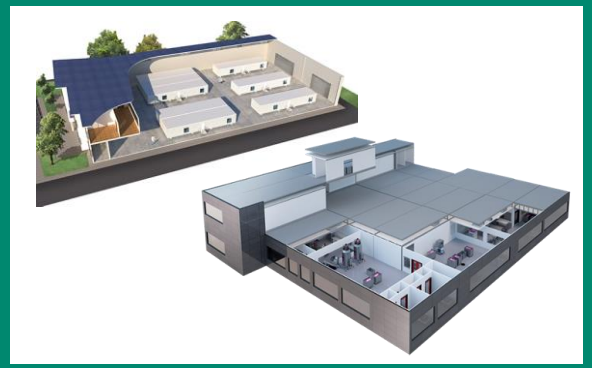
Facility investments, timing

Construction and validation costs

Manufacturing costs

How are we addressing current and future needs?

Facility



Single-use technology

Automation, process control, and digitalization

Process intensification

What is next-generation processing?

Intensified process

Continuous manufacturing

Process intensification of existing bottlenecks

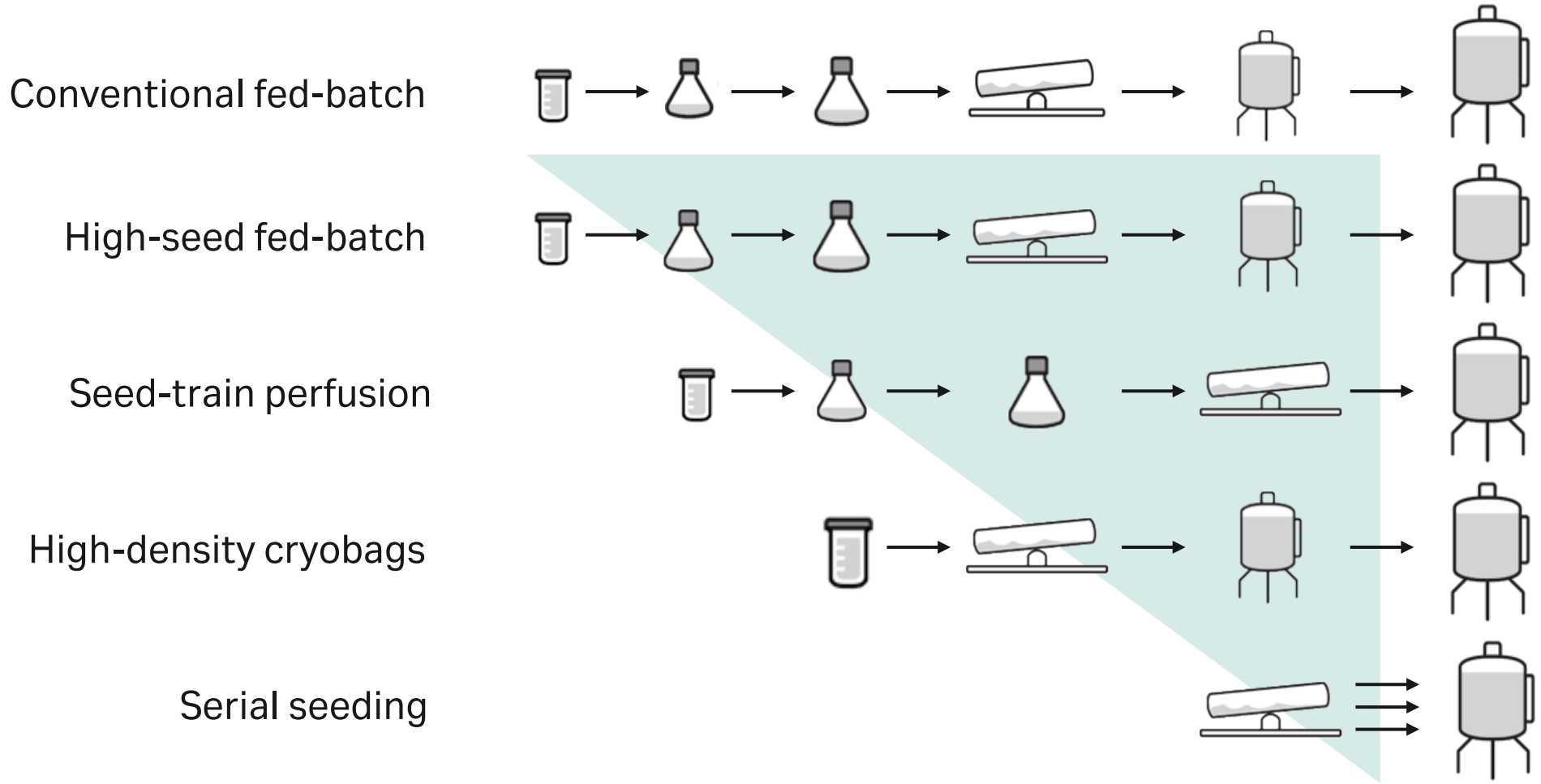
Solutions:

- Perfusion media development
- Intensified seed train
- Intensified production bioreactor
- Intensified capture
- Inline conditioning/dilution
- Inline viral inactivation
- Flow-through polishing
- Inline virus removal
- Single-pass formulation

2

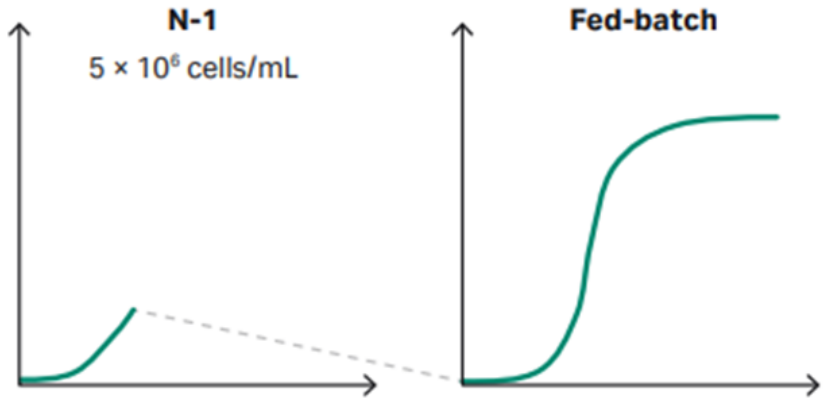
Case study: N-1 seed perfusion for fed-batch production process

Seed train perfusion can reduce time and/or bioreactor size



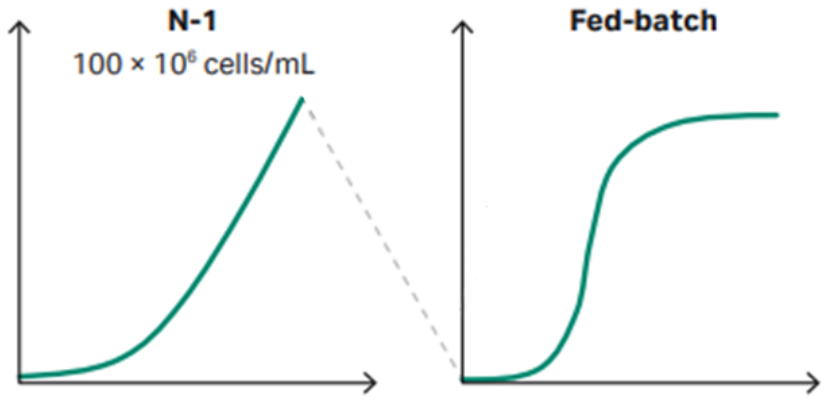
Up to 20-fold greater seed scale-up

Conventional fed batch cell growth



N-1 working volume	Seed volume at 0.5 MVC/mL
5 L	50 L
25 L	250 L
50 L	500 L

High-density perfusion cell growth



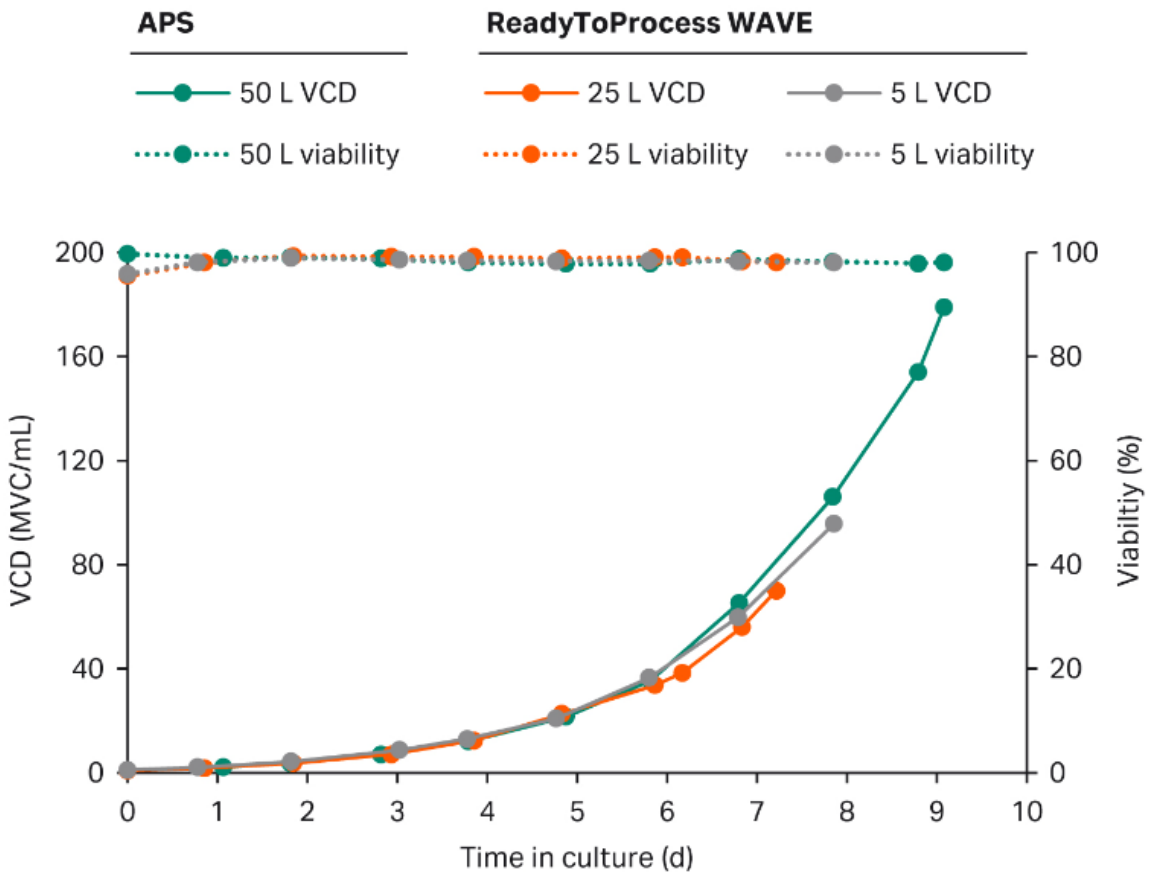
N-1 working volume	Seed volume at 0.5 MVC/mL
5 L	1000 L
25 L	5000 L
50 L	10 000 L

N-1 perfusion set-up using Xcellerex™ APS and XDR-50

Objective

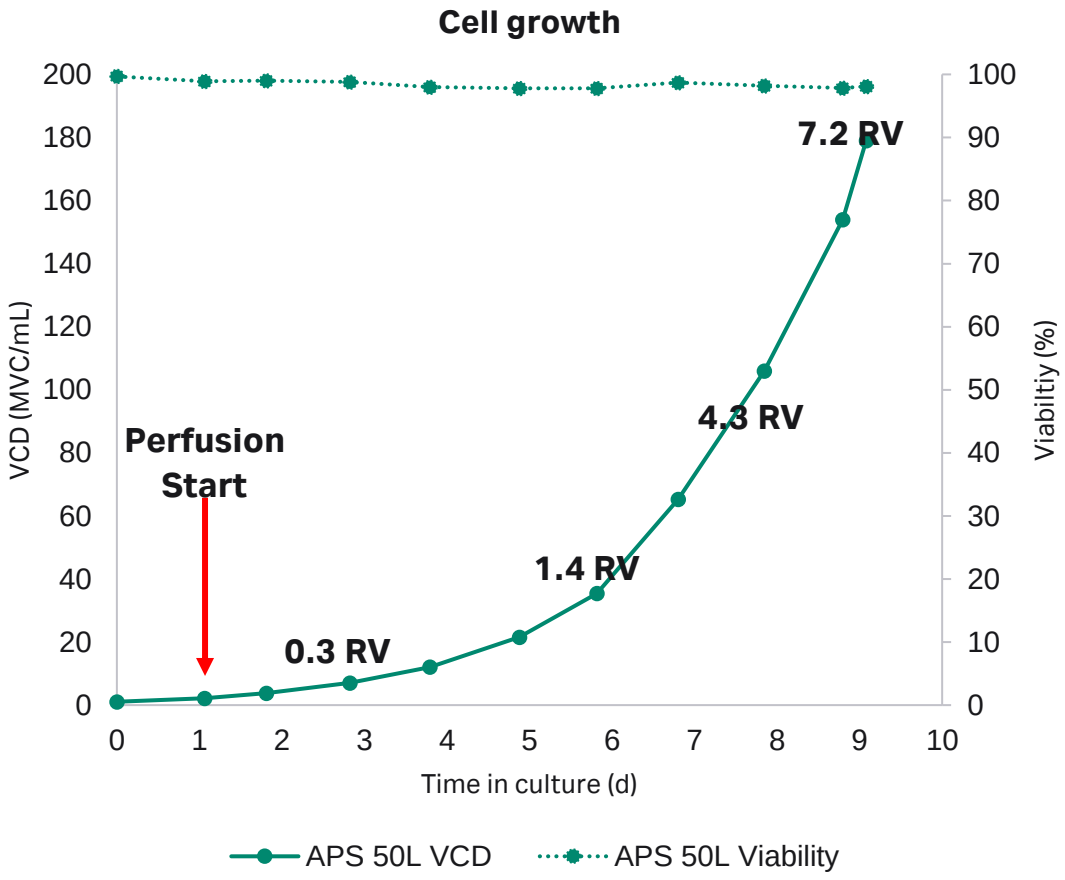
- Establish a CSPR based N-1 perfusion process targeting at least 100 MVC/mL in ReadyToProcess WAVE™ 25
- Maintain cells in exponential growth with a cell viability above 95%
- Scale the process to XDR-50 using Xcellerex™ APS
- Compare growth and productivity of production cultures seeded with N-1 perfused cells and with a conventional seed train

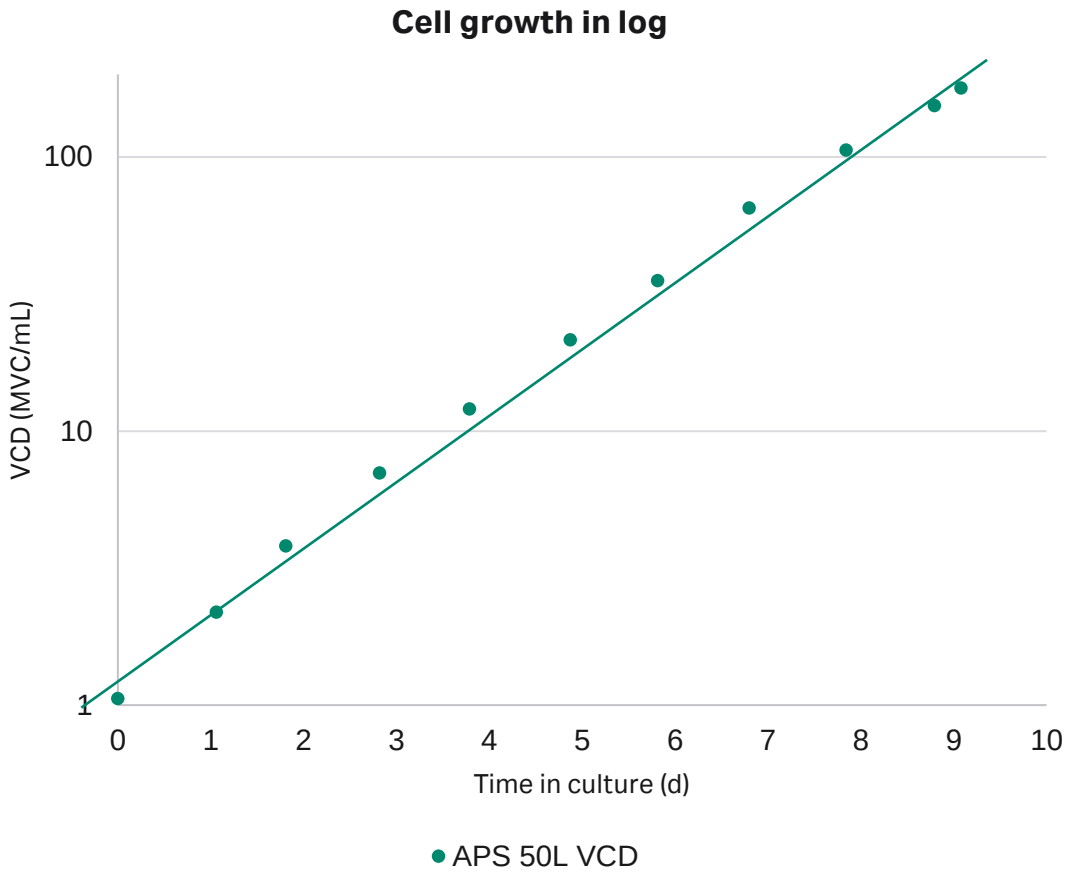
Perfusion N-1 Seed - ReadyToProcess WAVE™ 25 vs APS XDR-50



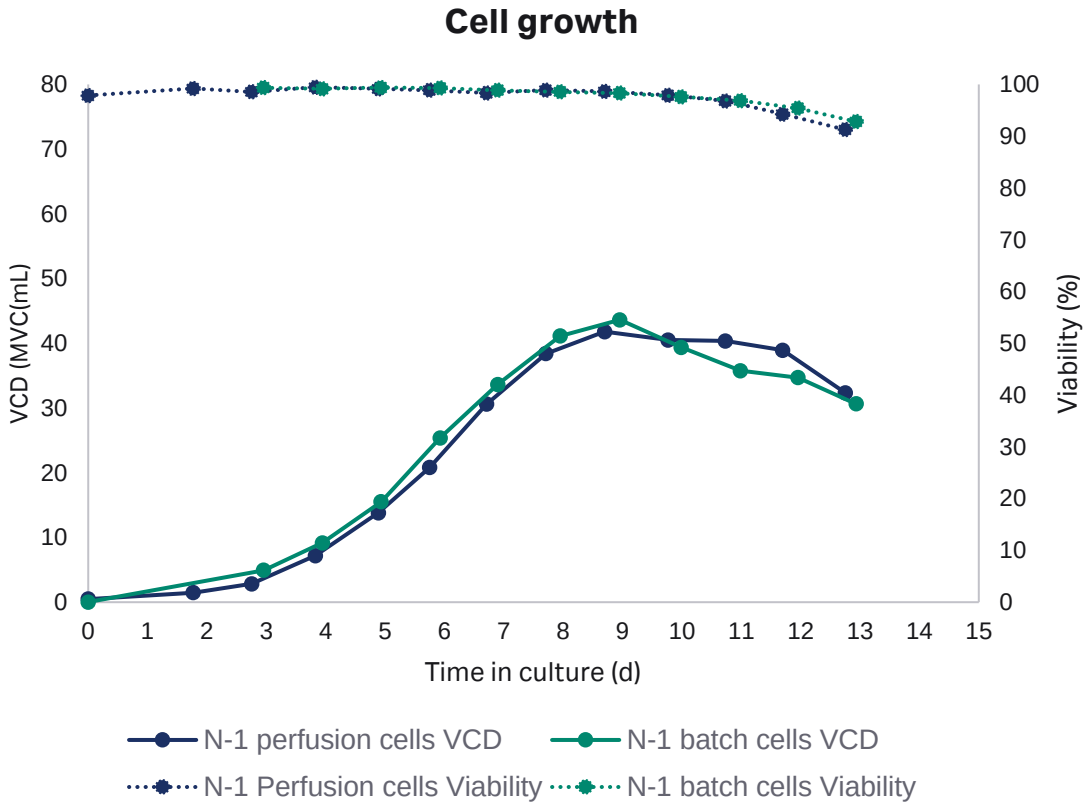
Effective scale-down of perfusion seed process using ReadyToProcess WAVE™ 25 for process development scale. Representative of perfusion seed performance in Xcellerex™ XDR-50 with APS

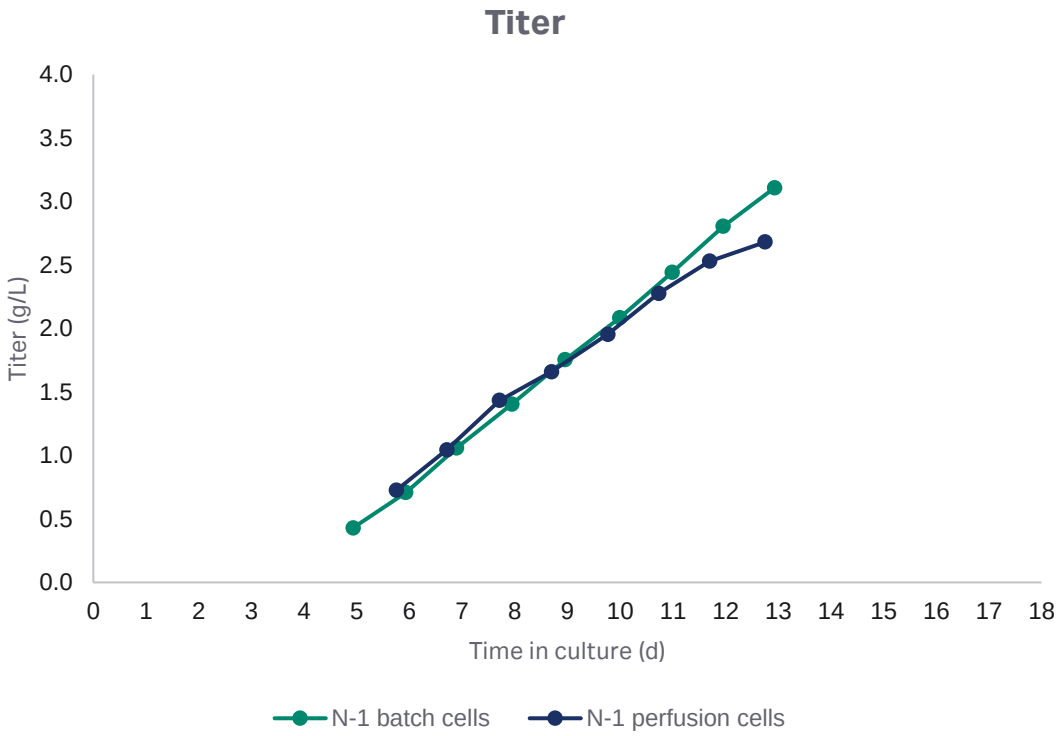
Seed growth to 179 MVC/mL in Xcellerex™ XDR-50 with APS





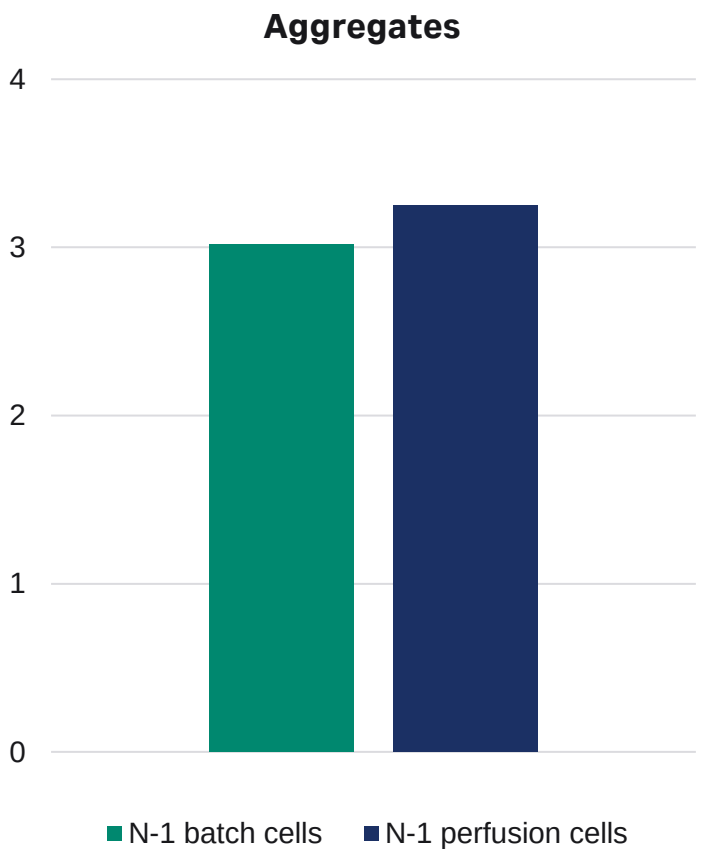
Comparable growth and productivity in fed-batch using N-1 inoculum

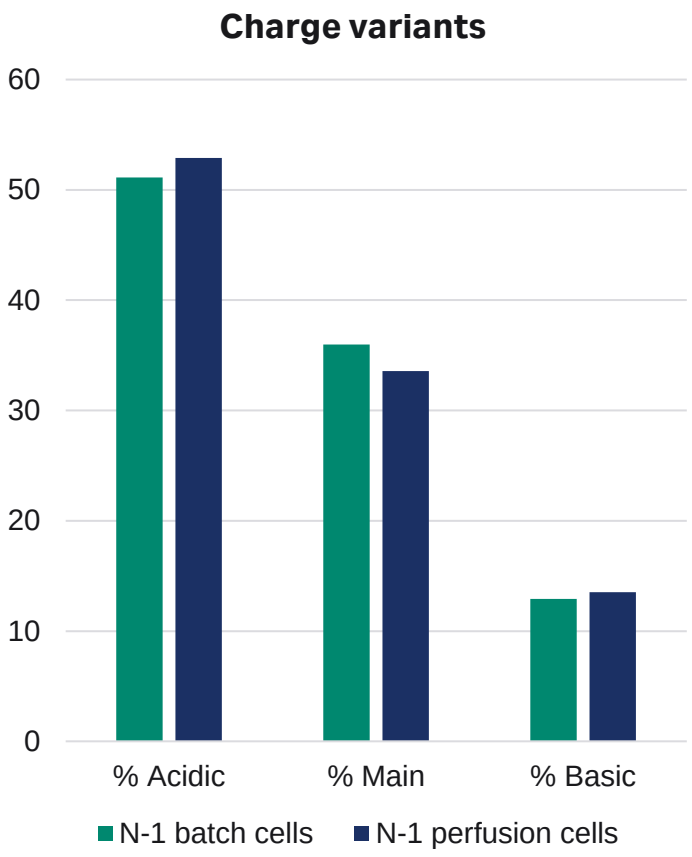


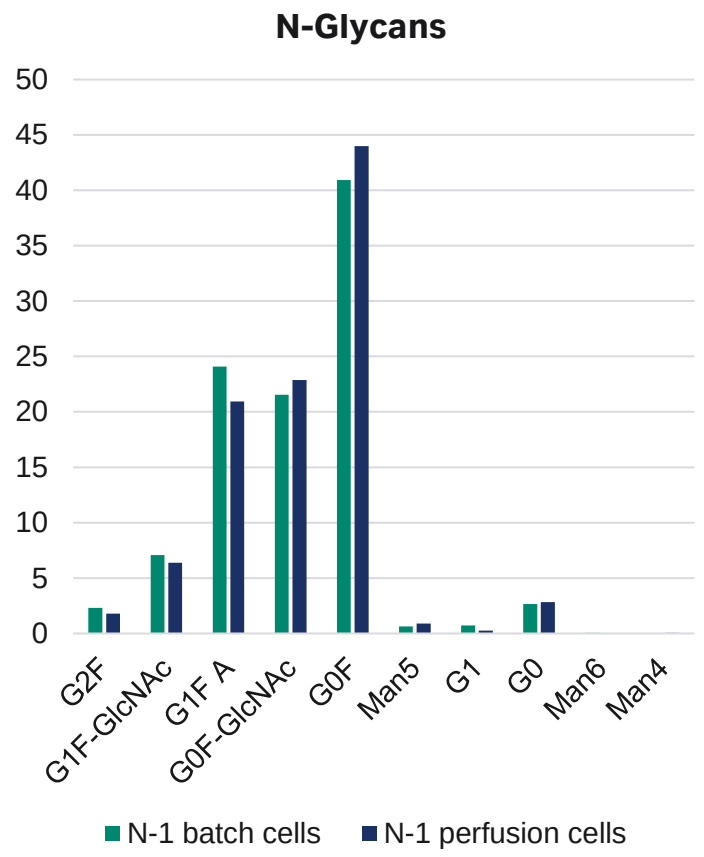


N-1 perfusion seed (179 MVC/mL) and N-1 batch seed as inoculum for fed-batch production culture

N-1 perfusion does not affect final product quality attributes







Conclusion

- Successful N-1 perfusion run in the XDR-50 using the Xcellerex™ APS
- We were able to achieve exponential growth to 179 MVC/mL and a cell viability over 95%.
  - An improvement in terminal seed densities of traditional methods - batch (5 MCV/mL) and rocking bioreactor perfusion methods (50-70 MVC/mL).
  - Processing to higher densities shorten time to production inoculation or allow increase in production process volume.
- ReadyToProcess WAVE™ 25 perfusion process scale in growth profiles with Xcellerex™ APS/XDR-50.
- Resulting production fed-batch process indicate equivalent performance between WAVE™ 25 an Xcellerex™ APS N-1 inoculum.
- Comparison of N-1 perfusion seed trains indicate consistent behavior between product platforms for the growth of both the seed and the subsequent production fed-batch cultures.
- Processing of the final protein of the production fed-batch cultures confirmed consistent protein titers and resulting product quality profiles regardless of seed source.

3

Bring consistency to perfusion

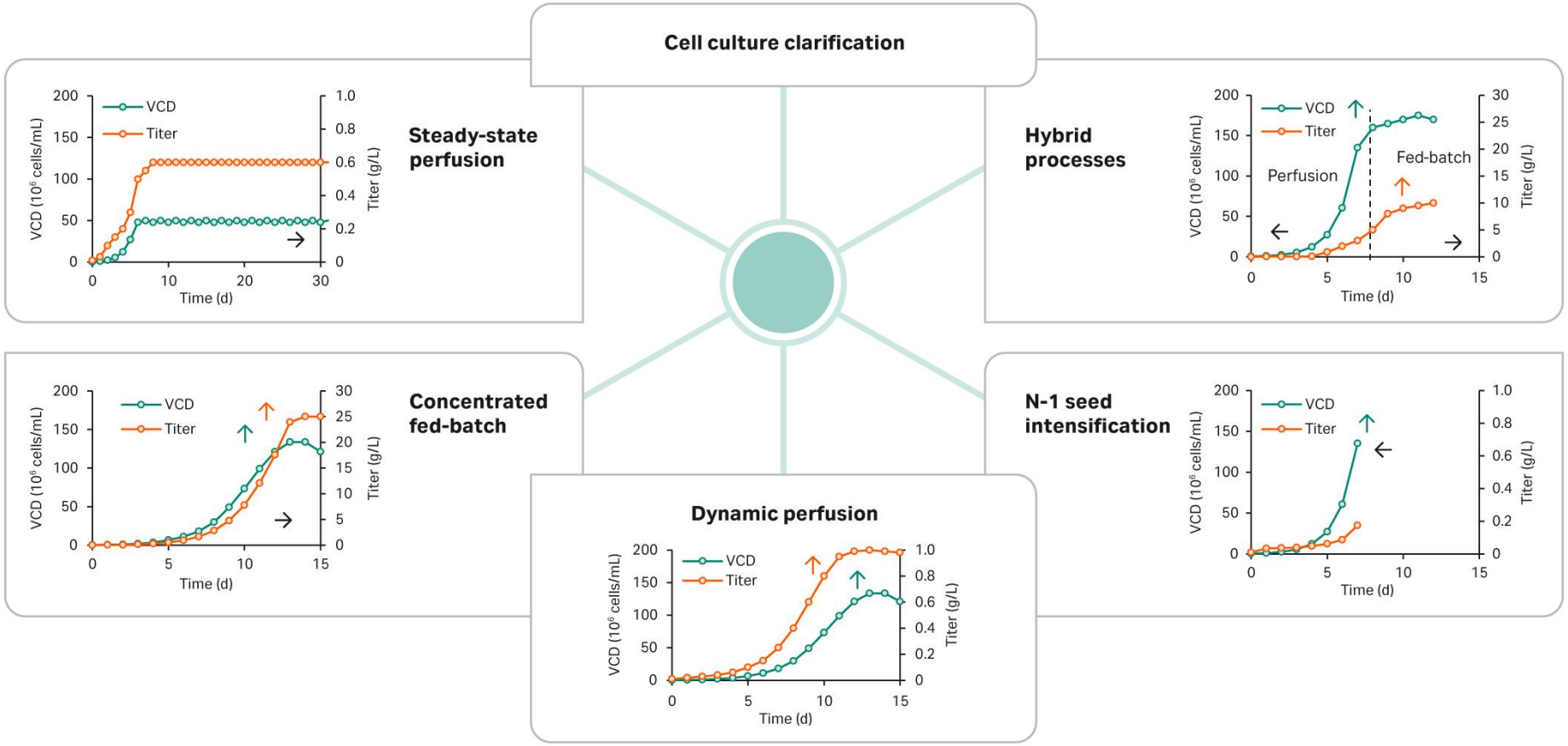
Introducing Xcellerex™ APS

Automate perfusion and reduce risk while ensuring that your process needs are met

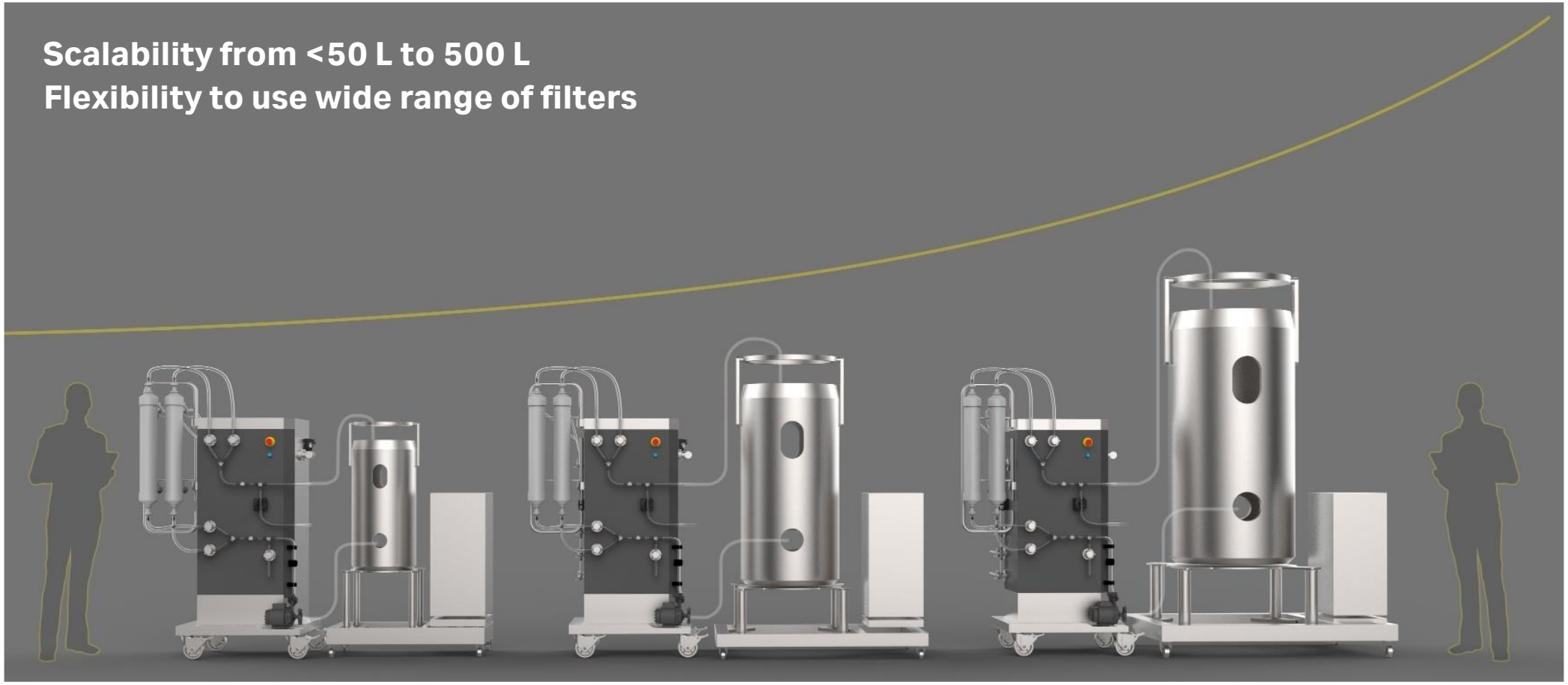
Xcellerex™ APS single-use perfusion system

- Automates upstream process intensification
- Minimizes opportunities for operator errors and lost batches
- Integrates Xcellerex™ bioreactor, automation, sensors, single-use flow kit, hollow fiber filters, and 3D bags
- Intuitive, scalable, single-use perfusion system

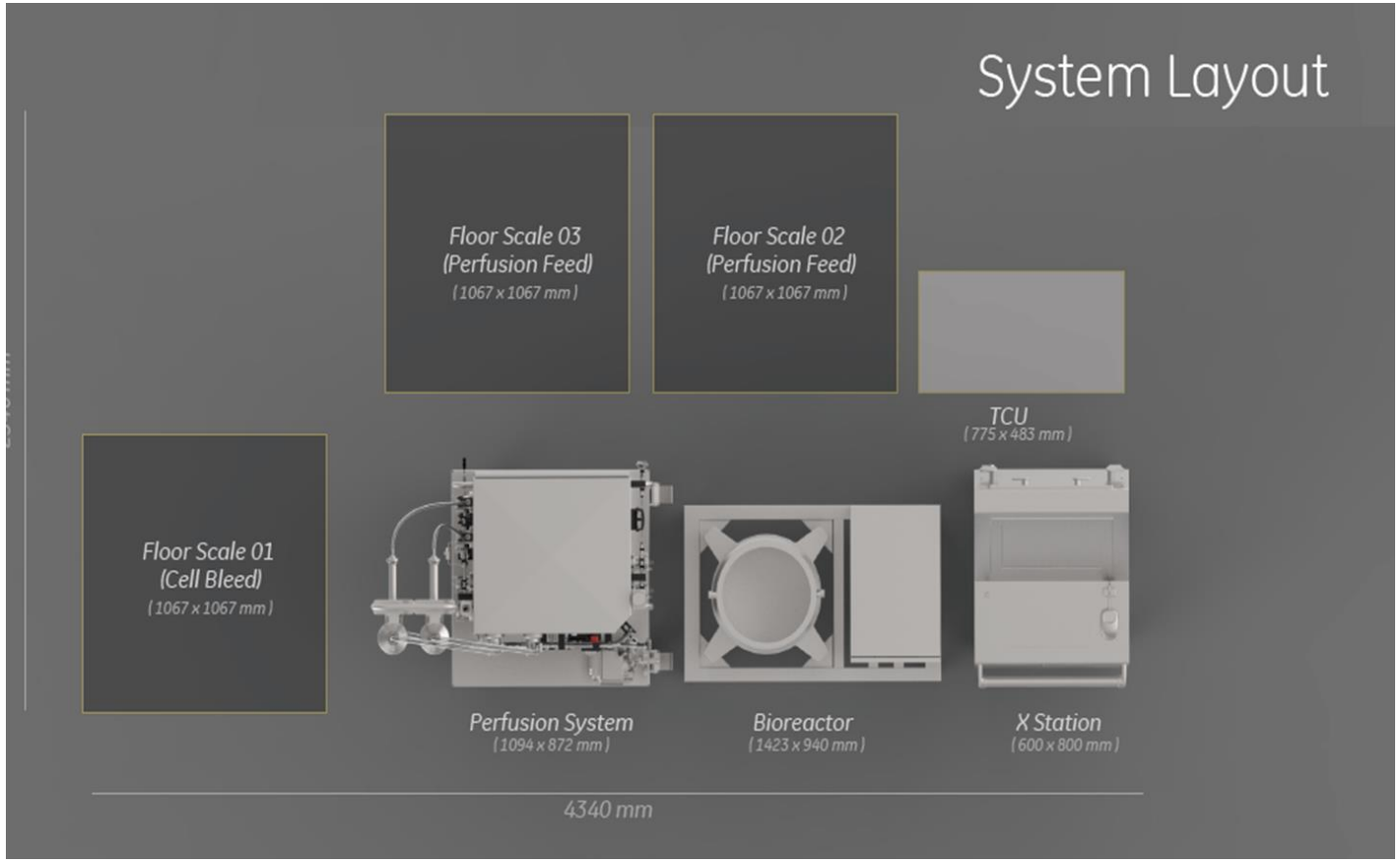
Use the same system for multiple applications in both pilot- and manufacturing-scale GMP operations



Broad range of operation



Compact, fully integrated perfusion operation



- Accurate: use floor scales to provide gravimetric control
- Continuous operation: perfusate can be fed directly to the protein A step

Simplifying perfusion operations

Intelligent filter switch

Automated liquid management

Connection between Xcellerex™ APS and bioreactor

Single-use flow path installation

Single user interface

Multiple perfusion modes

Automated cell bleed

Cell harvest

Process simplicity using Xcellerex™ APS Flow kit

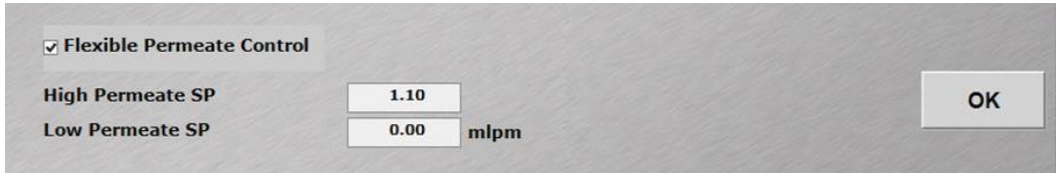
- Rapid start-up: flow kit sections assembled in less than 15 minutes
- Simplicity: back-up filter can be added in seconds and at any stage during the cell culture run
- Minimized bioburden: closed loop, presterilized, single-use flow path
- Complete process monitoring: precalibrated feed, retentate, and permeate pressure flow, as well as two flow sensors
- Minimized shear on cells: through smart design that avoids sharp bends in the flow path

Say goodbye to filter blockage interruptions

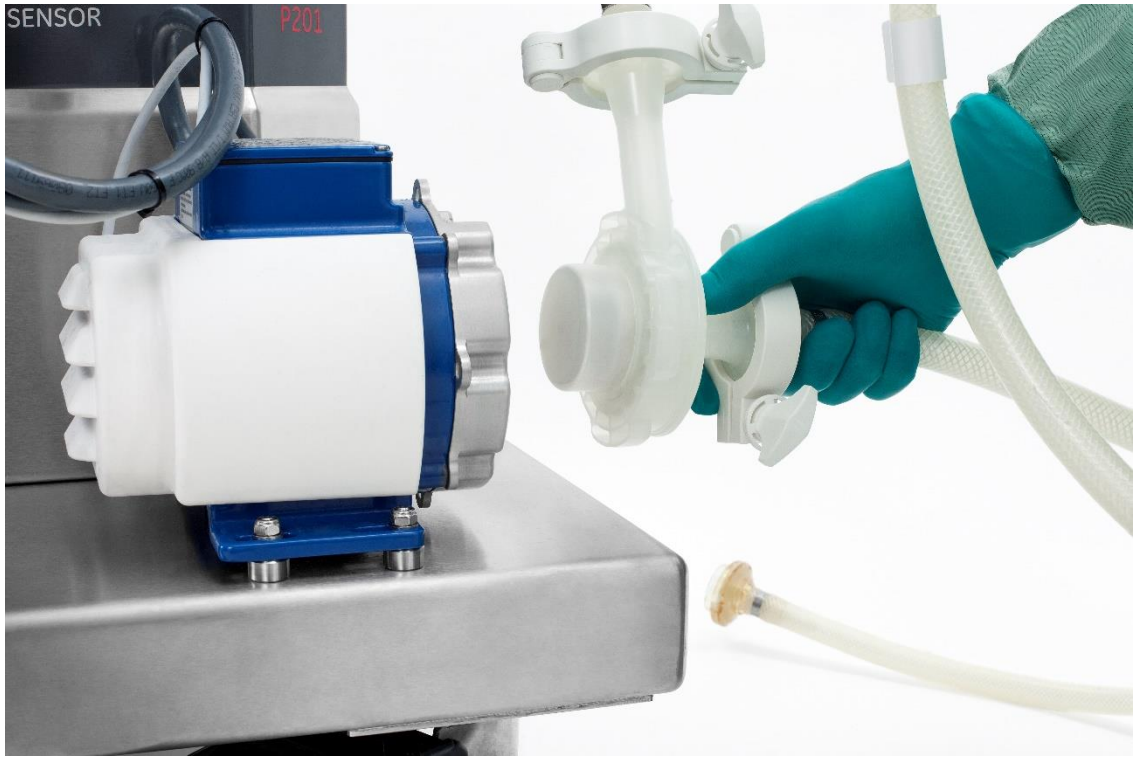
- Unattended operation: automated back-up filter switching
- No need for a second system: back-up filter to prolong process duration
  - Early prediction of filter fouling with key process parameters
  - Series of noncontact pinch valves to direct flow

Automated perfusion control and process monitoring

- Maintained steady-state throughout the process thanks to integrated floor scales (cell bleed, bolus feed addition, evaporation, etc.)
- Predictable and reliable processing through a weight-based approach that ensures a very tight process control
- Extended filter life through flexible permeate control



Low shear operation to maintain cell culture performance



- Utilizes the Levitronix™ single-use maglev centrifugal pump
- Proven performance in perfusion applications
- Handles high-viscosity solutions – ideal for high cell density applications
- Flow path designed to create minimal turbulence

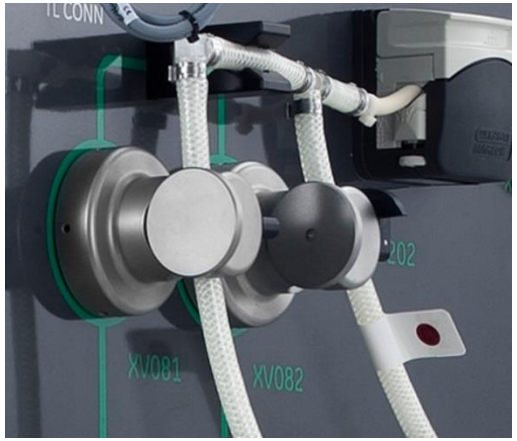
For details see:

Wang S, Godfrey S, Ravikrishnan J, Lin H, Vogel J, Coffman J. Shear contributions to cell culture performance and product recovery in ATF and TFF perfusion systems. *J. Biotechnol.* 2017;246:52-60. http://dx.doi.org/10.1016/j.jbiotec.2017.01.020

Key automation features for enhancing process control and cell growth

Automated medium bin switching

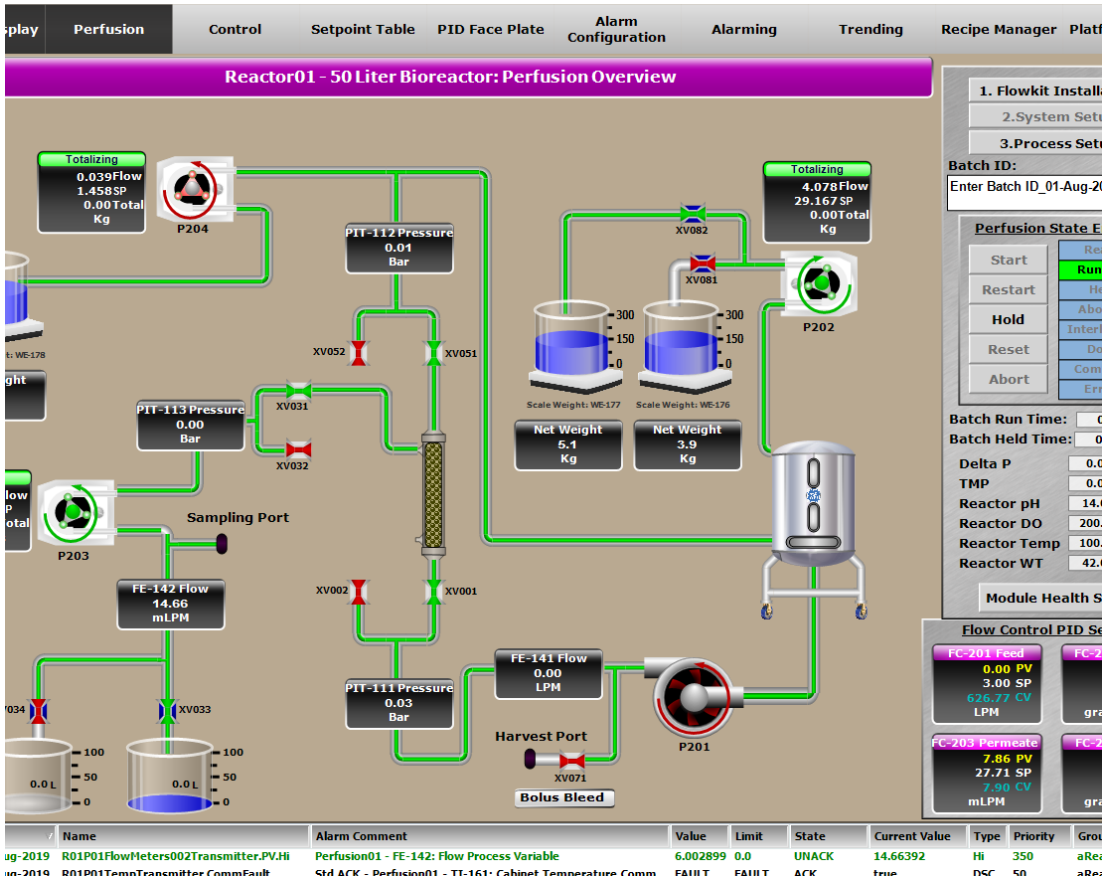
- Continuous, unattended cell culture medium addition into the bioreactor bag
- Automatically switch to new bin thanks to weight trigger



Automated cell bleed

- Maintain constant bioreactor cell density through automated cell bleed
- Cells can be bled from the bioreactor continuously or in bolus, enabling control of the cell density and cell growth

Fully integrated automation as standard



- Seamless integration with Xcellerex™ XDR bioreactors
- Simplified reporting: combined batch record for bioreactor and perfusion system
- Full monitoring and control with an interactive process layout

4

Summary

Bring consistency and flexibility to perfusion

- **Versatile**
  Flexible choice of hollow fiber filters, bioreactor sizes, accessories, and control options for efficiency.
- **Efficient**
  One automation platform and single user interface for Xcellerex™ bioreactors and Xcellerex™ APS supports traceability.
- **Productive**
  Easy filter change-over, liquid management switch, and cell bleed enhance process control and accuracy. This supports maintaining high cell densities with high cell viability, which boost productivity.

Designed for high cell density needs

- Bioreactor sizes up to 500 L
- Recirculation flow rate of 18 L/min max
- Hollow fiber cartridges micro/ultra, up to 4.4 m^2
- VVD of 0.075 to 2.0
- CSPR of 20 to 50 pL/cells/d

CSPR = cell specific perfusion rate
VVD = volume of fresh medium/working volume of reactor/day

Call to action

Do you want to see what APS can do for your process?

- Schedule a demo
- Visit Cytiva website for more information on Xcellerex APS
- Read the intensified seed culture for a fed-batch process Application Note
- Contact your Cytiva representative for more information

Thank you

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