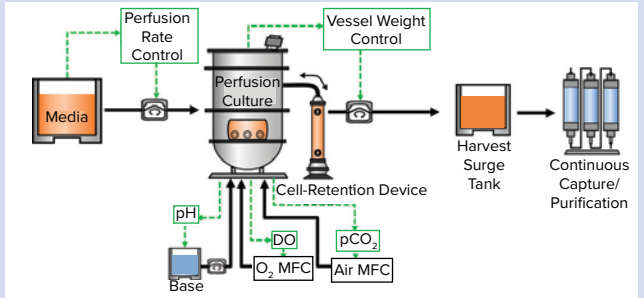
Continuous Biomanufacturing Implementation

Using an Intensified and Integrated Bioprocess Platform

Recent world events have demonstrated now more than ever the growing demand for pharmaceutical biologics that can be made rapidly and in high volumes yet somehow remain affordable. Hence, there is an urgent need to develop a next-generation biomanufacturing solution that provides high-yield, high-quality drug products and is highly flexible and cost-effective. Herein we describe the WuXi Biologics ultrahigh productivity platform (WuXiUP), an intensified perfusion culture process developed to meet the aforementioned need. WuXiUP adopts process-intensification strategies on to traditional perfusion culture processes to boost cell density and cell-specific productivity. The continuous harvest reduces greatly the residence time for a product within a bioreactor, leading to more desirable product quality and facilitating integrated continuous bioprocessing.

A typical configuration of a WuXiUP platform comprises a perfusion bioreactor coupled with a cell-retention device, which provides harvest to a direct product capture/purification system continuously (Figure 1). Microfiltration hollow-fiber filters usually are adopted to retain cells, and alternative tangential-flow (ATF) filtration is used for continuous harvest of a target protein in spent media. It is critical during continuous cell culture to keep a steady culture weight. That can be accomplished by monitoring and regulating both medium feed rate and harvest rate by means of feedback control to preselected target values. A multicolumn chromatography system that operates in a periodic countercurrent mode is applied to capture clarified harvest with three or more columns. For monoclonal antibodies (MAbs), protein A resin typically is used in such columns. For other molecule types, resin selection depends on the properties of both the resin and the harvest matrix. A multicolumn chromatography system captures harvested material continuously. Two columns are connected in tandem for feeding, and other column(s) are operated in parallel for product recovery, clean-in-place (CIP), and column-regeneration purposes.

Figure 1: Typical configuration of WuXiUP platform



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To demonstrate the performance of WuXiUP in producing biologics, a Chinese hamster ovary (CHO) cell line expressing an Fc-fusion protein was used for process evaluation. In this case, the WuXiUP process was compared with traditional fed-batch (TFB) and traditional perfusion

Figure 2: Cell-culture performance in different culture modes for an Fc-fusion protein producing CHO cell line: (A) profile of viable cell density (VCD) over 40 culture days, and (B) profile of accumulated Pv over 40 culture days

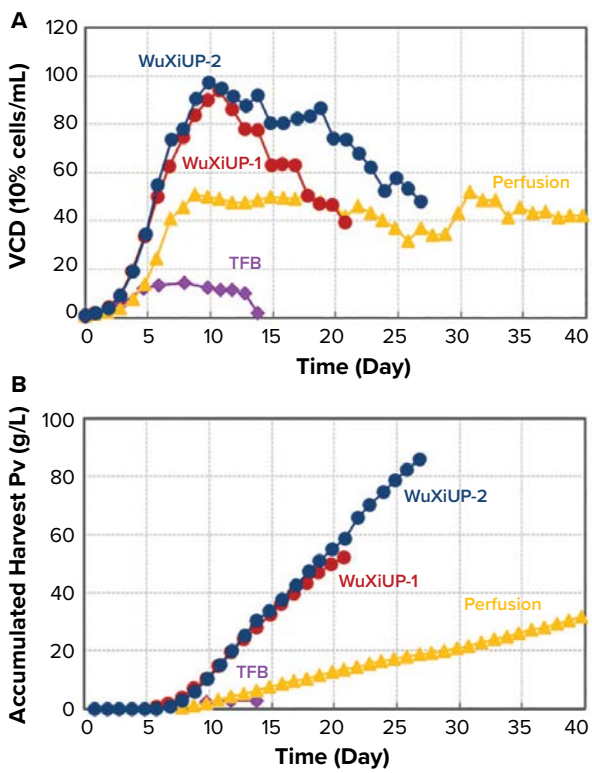


Figure 3: Pv comparison between WuXiUP and traditional fed-batch (TFB) in different projects

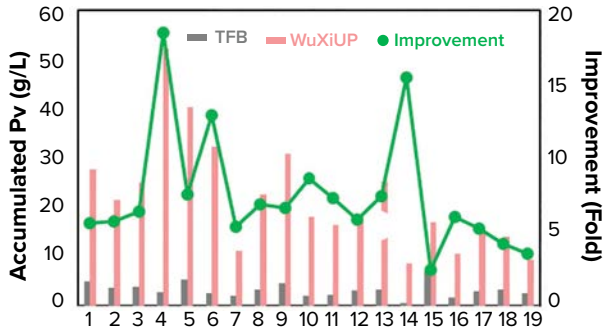


Table 1: Comparison of WuXiUP to standard bioprocess platforms

Process	Culture Duration (Days)	Accumulated Harvest Pv (g/L)	Daily Productivity (g/L per day)
TFB	14	2.82	0.20
Perfusion	40	31.63	0.79
WuXiUP-1	21	52.14	2.48
WuXiUP-2	27	85.86	3.18

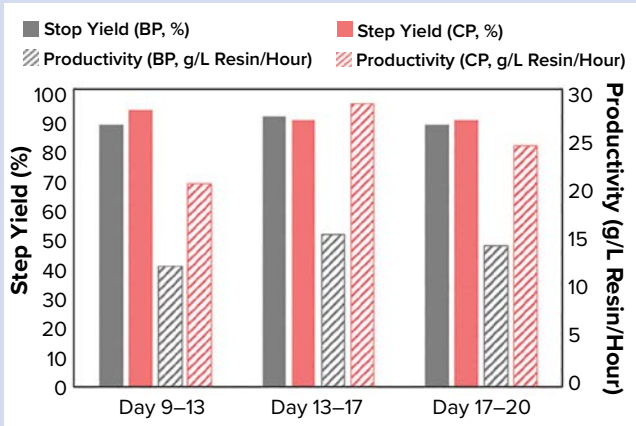
culture modes. Temperature-shift strategies were used for WuXiUP and TFB when the viable-cell densities (VCDs) approached their peak values. For the perfusion process, a target VCD of 50 × 10^6 cells/mL was set and maintained by adjusting the cell-bleeding rate. Figure 2A shows growth profiles. A significant boost in biomass accumulation was observed with the WuXiUP process, but the perfusion process maintained a relatively stable cell-density profile.

Figure 2B shows trends in accumulated Pv along the time course for different processes. An exceedingly higher productivity was achieved within a much shorter culture duration for the WuXiUP-1 process (21 days) compared with that of the traditional perfusion culture (14 days). Final accumulated Pv for the WuXiUP-1 process was 52.14 g/L, which is 18.49-fold and 1.39-fold higher than in that of the TFB (2.82 g/L) and perfusion (37.55 g/L) processes, respectively.

Taking culture duration into consideration, WuXiUP-1 remained the best for daily productivity (2.48 g/L per day), which was 12.4-fold and 3.14-fold higher than of the TFB (0.20 g/L per day) and traditional perfusion processes (0.79 g/L per day), respectively. With process optimization, the WuXiUP-1 process was ungraded to the WuXiUP-2 process, which resulted in dramatically higher final Pv of 85.86 g/L, and daily productivity of 3.18 g/L per day, which supports use of WuXiUP as a platform for generating different types of pharmaceutical proteins with extremely high productivity. The ultrahigh productivity and relatively short culture duration clearly demonstrates why WuXiUP is considered an intensified perfusion culture process.

In addition to the proof-of-concept study outlined above, WuXiUP has been applied to multiple projects, thus providing comprehensive evaluation of this platform technology. To demonstrate the plug-and-play feasibility of the platform,

Figure 4: Step yield and productivity comparison between batch-mode capture processes (BP) and continuous capture process (CP).



more than 15 clones were evaluated using the WuXiUP process. On average, a 7.4-fold yield improvement and a 4.5-fold increase in daily productivity were achieved over the TFB mode (Figure 3). The WuXiUP platform enables almost any biologic (e.g., MAbs, fusion proteins, and recombinant proteins) to be manufactured at ultrahigh productivity.

Downstream productivity of the capture step was doubled with application of continuous capture technology, and comparable process performance was obtained compared with a traditional batch process (Figure 4). With higher productivity, the resin amount, buffer consumption, and facility footprint were reduced significantly, Such factors help reduce costs dramatically on a batch-production basis. In addition, the combination of an intensified perfusion culture process and continuous-capture process has been successfully scaled up to 200-L scale in a good manufacturing practice (GMP) facility.

As an integrated, intensified, continuous biologics manufacturing technology platform, WuXiUP is driving improved economics, flexibility, and quality. This remarkably scalable and robust cell-culture process can be developed rapidly or converted from a traditional fed-batch process with five- to 10-fold higher productivity. This exciting new technology enables 1,000–2,000-L disposable bioreactors to achieve similar productivity as traditional 10,000–20,000-L stainless-steel bioreactors. Thus, WuXiUP can expedite product launch while lowering a company's capital and operating costs and achieving similar or better product quality than traditional processes.

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